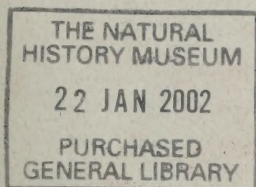


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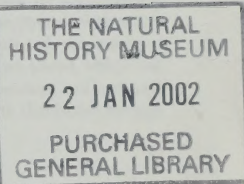
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Dr. Sicard's tenure will begin with the 2002 volume (63) of JKAS. Papers being submitted for publication in the journal should now be sent directly to him.

The current editor, Dr. John W. Thieret, Northern Kentucky University, expresses his indebtedness to the many authors who have contributed papers to the journal during his several year editorship. He has special thanks for Dr. Robert J. Barney, who has been the editor of abstracts for JKAS, and to Varley Wiedeman, who has been the compiler of indexes for the journal.



## **Suitability of Various Oak (*Quercus*) Species for Gypsy Moth (*Lymantria dispar*) Growth and Development**

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Department of Entomology, University of Kentucky, Lexington, Kentucky 40546-0091

### **ABSTRACT**

We determined the relative suitability of four oak (*Quercus*) species commonly found in urban landscapes for gypsy moth (*Lymantria dispar*) growth and development and attempted to characterize the physical basis for differences in host suitability. Of the oak species tested—burr oak (*Q. macrocarpa*), white oak (*Q. alba*), pin oak (*Q. palustris*), and willow oak (*Q. phellos*)—gypsy moth growth was highest on burr oak, intermediate on willow oak, and lowest on pin and white oaks. Caterpillar development time did not differ significantly among the species we tested. Caterpillar frass production, used as an indirect measure of leaf consumption, varied significantly among oak species, being greatest on willow oak and least on white oak. We also measured leaf toughness and leaf density. Burr oak and white oak had tougher, denser foliage than pin oak, with willow oak being intermediate. In spite of the tougher, denser foliage, caterpillars fed burr oak outperformed caterpillars fed the other oak species. However, our data suggest that of the species we tested, willow oak is likely to suffer the greatest defoliation. Our results will be useful to arborists, urban foresters, and landscape managers in planning long-range approaches to minimizing the impacts of gypsy moth defoliation through silvicultural techniques.

### **INTRODUCTION**

The gypsy moth (*Lymantria dispar*; Lepidoptera: Lymantriidae), an introduced defoliator of deciduous trees, has an extremely wide host range and the potential to reach outbreak levels (Drooz 1985). Since its introduction from Europe in the late 1800s, the moth has established throughout northeastern United States. Its continued spread across much of North America is assured by an abundance of several oak (*Quercus*) species, which are among the preferred host plants (Leibhold et al. 1995). Kentucky is on the leading edge of the expanding gypsy moth infestation in North America and is predicted to be generally infested by the year 2015 (U.S.D.A. Forest Service 1991).

The gypsy moth is univoltine and overwin-

ters in masses of 100 to 1000 eggs, which are typically found in bark crevices but may also be laid on automobiles, outdoor equipment, and other objects, thus facilitating human-assisted, long-distance dispersal (Drooz 1985). Eggs hatch in early spring, and young larvae frequently spin silken threads and can be transported short distances by the wind. Newly hatched larvae feed at the base of expanding leaves; older larvae feed on leaf edges and eventually consume entire leaves. Larvae pupate in sheltered locations and emerge as adults about 2 weeks later.

When gypsy moth populations are high, forest canopies can be completely defoliated. The consequences of repeated defoliation include reduced tree growth, mortality, shifts in species composition, and changes in nutrient cycling and wildlife distribution patterns (Doane and McManus 1981). Such defoliation can re-

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sult in loss of revenue due to reduced timber yield, oak mortality, quarantines, and declines in recreational use (U.S.D.A. Forest Service 1995).

Although long considered a major forest pest, the gypsy moth becomes an urban pest as populations become established in a region. The growing interface between urban and rural landscapes has resulted in increasingly visible acreage susceptible to devastation by gypsy moths. In addition to the tremendous defoliation potential, caterpillars are a nuisance and can be a human health hazard, causing allergic respiratory and dermal responses (Doane and McManus 1981; U.S.D.A. Forest Service 1995).

Arborists, urban foresters, and land managers in newly infested regions must make decisions regarding gypsy moth management that influence urban forest health, survival and appearance of street trees, and recreational and relaxation opportunities for the general public. Common control options in infested areas include applications of *Bacillus thuringiensis* var. *kurstaki* (Btk), a bacterium that produces a lepidopteran-specific toxin causing paralysis of the caterpillar gut; applications of insect growth regulators such as diflubenzuron; or applications of various conventional insecticides as foliar or trunk sprays (Shetlar and Herns 1999). The use of non-host trees in urban settings is a potential management strategy, but the positive attributes of many of our oak species make this a less appealing option.

The objective of our study was to determine the relative suitability of several oak species commonly found in urban landscapes for gypsy moth growth and development and to quantify some physical characteristics that might contribute to any differences in the results.

## MATERIALS AND METHODS

### Plant Material

We chose four oak species for our study. Burr oak (*Quercus macrocarpa*) is a drought-resistant, high-value component of deciduous forests and urban landscapes in central North America. White oak (*Q. alba*) is a major high-value component of deciduous forests and an important tree in urban areas throughout eastern U.S. Pin oak (*Q. palustris*) is a fast-growing species of the central hardwoods region

that is particularly important in urban landscapes as a street and landscape tree. Willow oak (*Q. phellos*), a fast-growing species of more southerly distribution, is important in both forest and urban landscapes and is widely planted as a street and landscape tree (Burns and Honkala 1990). Thus, we tested two major forest and green space species, burr and white oaks, and two major landscape and street tree species, pin and willow oaks.

Plant material was purchased in spring 1997 as 2-yr-old whips from commercial nurseries and planted on the University of Kentucky's Spindletop Research Farm in a randomized block design with six blocks. Fertilizer was applied as needed.

In late May 2000 we removed terminal foliage from the south side of the four species at the mid-crown level. We used foliage from trees in four blocks for the insect assays and foliage from trees in all six blocks to measure foliar physical characteristics.

### Insect Assays

Gypsy moth caterpillar growth and development were monitored on foliage from each of the four oak species, which had been surface-sterilized by submerging in a 0.1% sodium hypochlorite solution for 5 min (Rieske and Raffa 1998). Gypsy moth larvae were obtained from the USDA-ARS laboratory (Otis Air Force Base, Massachusetts, USA) and held in growth chambers with a 15:9 (L:D) photoperiod at 23°C in the University of Kentucky Forest Entomology Quarantine facility. Newly molted fourth instar caterpillars previously fed a wheat-germ-based artificial diet (Southland Products Inc., Lake Village, Arkansas) were starved for 24 hr prior to use in assays.

We assessed caterpillar performance by allowing larvae to feed for the duration of the fourth stadium. Foliage from each oak species was placed in florists' water picks in 7 × 21 cm clear plastic rearing boxes. Fourth instar caterpillars were individually placed on foliage and monitored at 24 hr intervals. Three insects were used for each of the four tree species, replicated over four blocks, for a total of 48 assays. Leaves were replaced at 2–3 d intervals to ensure freshness. Immediately after molting, insects were removed from the foliage and frozen. At the completion of the assay, plant tissue, insect cadavers, and waste mate-



rial were oven dried at 60°C for 10 d and weighed. Relative growth rate (RGR = caterpillar biomass gained [mg]/initial caterpillar wt [mg]/time [d] and length of caterpillar stadium (duration of 4th instar in d) were calculated as measures of caterpillar performance and foliar suitability.

### Foliar Characteristics

Leaves collected concurrently for fresh weight analysis were excised at the leaf base, and foliar leaf area (LI-3100 Area Meter, LiCor, Inc., Lincoln, Nebraska) and fresh weight immediately measured. We calculated leaf density (g/cm<sup>2</sup>) and measured leaf toughness from three leaves per tree for each of the six trees per species (N = 18 per oak species) to determine whether caterpillar performance could be correlated with foliar physical characteristics. We determined foliar toughness by measuring the force (kg) necessary to penetrate leaf tissue using a force gauge fitted with a pointed cone attachment (Mark 10 Corp., Hicksville, New York).

### Statistical Analysis

We used analysis of variance with a randomized block design to detect differences in caterpillar growth and development and in foliar characteristics among the oak species. When necessary, data were log transformed to adjust for heterogeneous variances, and significant differences were assessed using Fisher's protected LSD. We assessed correlations between caterpillar performance and foliar physical characteristics.

## RESULTS

### Insect Assays

The relative growth rate of gypsy moth caterpillars was significantly affected by host species ( $F_{3,9} = 5.396$ ,  $P < 0.05$ ). Caterpillar relative growth rate was greatest on burr oak and was significantly higher than the growth rate of caterpillars fed pin and white oaks (Figure 1A). Caterpillar growth rate on willow oak was intermediate and not significantly different than the growth rate on burr oak or the growth rate on pin and white oaks.

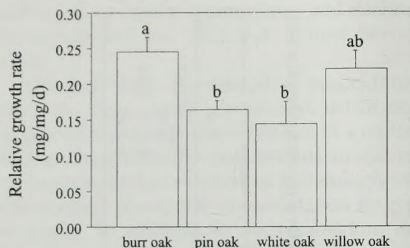
Gypsy moth development time was not significantly affected by host species ( $F_{3,9} = 2.936$ ,  $P > 0.05$ ). Although development time was quickest for caterpillars fed burr oak (Fig-

Table 1. Foliar characteristics (mean  $\pm$  SE) of four oak species from Spindletop Research Farm, University of Kentucky, assayed for gypsy moth caterpillar host suitability, May 2000. Means within rows followed by the same letter are not significantly different.

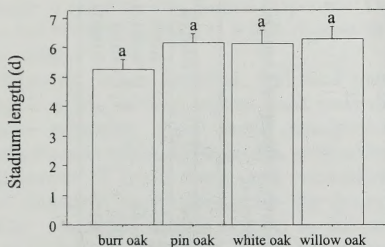
Foliar characteristic	Species				$F_{3,15}$ $P$
	Burr oak	Pin oak	White oak	Willow oak	
Leaf toughness (kg)	0.034 $\pm$	0.031 $\pm$	0.036 $\pm$	0.034 $\pm$	$F_{3,15} = 3.2P$ < 0.05
Foliar density (g/cm <sup>2</sup> )	0.001 a	0.001 b	0.002 a	0.001 ab	$F_{3,15} = 4.5P$ < 0.05
	0.012 $\pm$	0.010 $\pm$	0.012 $\pm$	0.011 $\pm$	
	0.0004 ab	0.0003 c	0.0010 a	0.0003 bc	



## A) Relative growth rate



## B) Stadium length



## C) Frass production

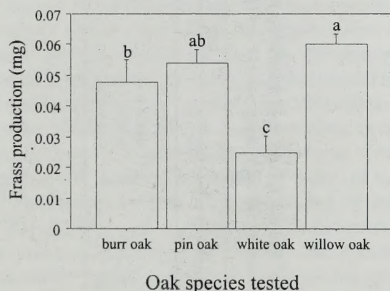


Figure 1. Performance of gypsy moth caterpillars fed four species of oaks from Spindletop Research Farm, University of Kentucky, May 2000. Caterpillar (A) growth rate, (B) development time, and (C) frass production. Means followed by the same letter are not significantly different ( $P < 0.05$ ).

ure 1B), there were no significant differences in stadium length among the oak species we tested.

Although we did not calculate caterpillar consumption rates, we measured the amount of frass produced, which is indicative of consumption, by insects reared on each oak species. Frass production was significantly affect-

ed by host species ( $F_{3,9} = 11.868$ ,  $P < 0.01$ ). Caterpillars fed willow oak produced significantly greater amounts of frass than did those fed burr or white oaks (Figure 1C); those fed pin oak produced frass in quantities similar to willow and burr oaks.

## Foliar Characteristics

Leaf toughness varied among species and was greatest in burr and white oaks, and lowest in pin oak (Table 1,  $F_{3,15} = 3.198$ ,  $P < 0.05$ ). Willow oak leaf toughness was intermediate and not significantly different from the other species. Leaf density was also species dependent (Table 1,  $F_{3,15} = 4.473$ ,  $P < 0.05$ ) and was greatest for burr and white oaks. Again, leaf density of willow oak was intermediate, and there was no significant difference in leaf density between burr and willow oaks, and no significant difference between willow and pin oaks.

Caterpillar growth rate was negatively and significantly correlated with development time; insects that grew faster also grew larger ( $r = -0.432$ ,  $Z = -3.031$ ,  $P = 0.0024$ ). Leaf toughness and leaf density were also strongly correlated ( $r = 0.486$ ,  $Z = 4.374$ ,  $P < 0.001$ ). However, there were no significant correlations between insect growth rate and leaf toughness ( $r = 0.035$ ,  $Z = 0.225$ ,  $P = 0.8221$ ) or between insect growth rate and leaf density ( $r = -0.014$ ,  $Z = -0.089$ ,  $P = 0.9293$ ). Similarly, there were no significant correlations between insect development time and leaf toughness ( $r = -0.026$ ,  $Z = -0.167$ ,  $P = 0.8670$ ) or between insect development time and leaf density ( $r = -0.038$ ,  $Z = -0.247$ ,  $P = 0.8050$ ).

## DISCUSSION

Large size and rapid development are generally considered advantageous in insects (Price 1997). Larger caterpillars tend to develop into larger pupae; in the gypsy moth, pupal weights are directly correlated with fecundity (Drooz 1985). Thus larger insects produce more offspring and are therefore more successful than their smaller counterparts. Additionally, accelerated larval development time is advantageous since it allows for avoidance of natural enemies and other mortality factors.

Of the four oaks we tested, caterpillars fed burr and willow oaks had the highest growth

rates and produced relatively large amounts of frass, suggesting that these oaks are the most suitable for gypsy moth growth and development. Caterpillar relative growth rate was greatest, and development time was quickest, on burr oak. Caterpillars fed foliage from pin and white oaks had the lowest relative growth rates. Frass production was especially low from caterpillars fed white oak, indicating that, of the species we tested, this was the least suitable for gypsy moth development.

Surprisingly, the physical characteristics of leaf toughness and leaf density were highest on burr oak, the most suitable of host plants, contrary to predictions of host selection and suitability (Price 1997). We would have expected caterpillar consumption, and therefore growth rates, to be greater on tender foliage than on denser, tougher foliage. Although we did not measure relative consumption rates per se, we did measure the amount of frass produced, which is indicative of consumption. Frass production was greatest from caterpillars fed willow oak, which was among the least tough and dense. Therefore, our measure of caterpillar consumption was inversely related to leaf toughness, as we would predict. In spite of this, burr oak proved the most suitable host of the oak species tested for our caterpillars. However, if frass production is a measure of leaf consumption, our data suggest that, of the species we tested, willow oak is likely to suffer the greatest defoliation.

Host suitability for gypsy moths is affected by foliar quality, including defensive compounds (Schultz 1988) and nutritional components (Rieske and Raffa 1998; Roth et al. 1994). A variety of biological and environmental factors, including plant stress, can influence foliar quality. Trees in urban landscapes are subject to a variety of stresses, including drought, soil compaction, nutrient depletion, fungal infection, and attack by herbivorous insects, which may influence foliar chemistry. Our experimental trees may have been negatively impacted by the severe drought suffered throughout the region in 1999. We did not investigate the chemical attributes of the oak fo-

liage in our study, and the implications of our results are somewhat limited by the absence these data. Clearly these additional factors would contribute to host suitability for gypsy moth caterpillars.

#### ACKNOWLEDGMENTS

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## Impact of Feeding Behavior of Beavers (*Castor canadensis*) on Woody Plants at Owsley Fork Reservoir in Eastern Kentucky

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### ABSTRACT

The beaver (*Castor canadensis*) is a widely distributed, large rodent capable of causing considerable damage in loss of timber revenues. Beavers occur throughout Kentucky; however, no published data exist with regard to the habits of this species in the state. During January and February 2000, we sampled the woody plants damaged by recently colonizing populations of beaver at Owsley Fork Reservoir in eastern Kentucky and compared these data to woody plant species available in the habitat. Beavers were opportunistic in their foraging patterns, feeding upon 18 species of woody plants; however, they exhibited selectivity in diet choice ( $\chi^2 = 14.9$ ,  $P < 0.025$ ). No species of woody plant was preferentially damaged by beavers, but yellow-poplar (*Liriodendron tulipifera*) and oaks (*Quercus* spp.) were avoided. Species damaged most frequently included Virginia pine (*Pinus virginiana*), willow (*Salix* spp.), and American beech (*Fagus grandifolia*). Beavers concentrated their feeding activity within 4 m of the shore and damaged the 0-4 cm and the >4-14 cm diam size class of woody plants most frequently. These data suggest that beavers imparted minimal impact to existing timber resources, largely impacting unmerchantable species and selecting small diameter stems close to the shore.

### INTRODUCTION

Although beavers (*Castor canadensis*) are widely distributed throughout Kentucky (Barbour and Davis 1974), relatively little is known of their ecology in this state, especially the eastern Cumberland Plateau region. Once virtually eliminated by trapping and land-use changes, populations of beaver have recovered in Kentucky through restocking efforts and are now locally common and believed to be at pre-settlement densities (Barbour and Davis 1974). Beavers have been the subject of much investigation (for reviews see Hill 1982; Novak 1987), and studies of their diet, food preferences, and harvest of plants used in lodge construction exist for many portions of the range (e.g., Aleksiuk 1970; Barnes and Mallik 1996; Brenner 1962; Hall 1960; Nixon and Ely 1969; Northcott 1971; Pinkowski 1983; Roberts and Arner 1984; Shipps et al. 1979; Svendsen 1980). Foraging of beavers is consistent with models describing optimal foraging in herbivores (Belovsky 1984; Jenkins 1980; Pinkowski 1983), with the size-distance relationship, i.e., size of food item relative to distance from shore, reversed because of the large size of food items relative to the mass of the beaver (Fryxell and Doucet 1993; Schoener 1979). Beavers exhibit sampling behavior, i.e., con-

suming only a small portion of the available food item in a feeding bout, permitting them to assess temporal and spatial variation in food resources (Jenkins 1978), further maximizing their feeding efficiency.

Composition of the diet of beavers varies regionally, with aspen (*Populus tremuloides*) and alder (*Alnus* spp.) important in Newfoundland (Northcott 1971), sugar maple (*Acer saccharum*) and yellow birch (*Betula alleghaniensis*) in Michigan (Belovsky 1984), aspen and green ash (*Fraxinus pennsylvanica*) in North Dakota (Pinkowski 1983), and aspen and willow (*Salix* spp.) in California (Hall 1960). In the southeastern United States, beavers feed heavily on sweetgum (*Liquidambar styraciflua*) in South Carolina (Shipps et al. 1979) and sweetgum and loblolly pine (*Pinus taeda*) in Louisiana (Chabreck 1958). In the Appalachian mountain region, beavers consume aspen and red maple (*Acer rubrum*) in Pennsylvania (Brenner 1962), and yellow-poplar (*Liriodendron tulipifera*), American hornbeam (*Carpinus caroliniana*), alder (*Alnus serulata*), and maples (*Acer* spp.) in Ohio (Nixon and Ely 1969; Svendsen 1980).

Beavers are capable of causing significant economic loss by cutting and girdling trees and by creating impoundments that flood valuable timber resources (Novak 1987). An-



nual damage in the United States alone was estimated at \$75–\$100 million (Miller 1983), and economic loss over a 40-year period in the southeastern United States was estimated to be \$4 billion (Arner and Dubose 1982). Diet of beavers in adjacent Ohio include timber species, such as yellow-poplar (Svendsen 1980), that are a highly valued resource in Kentucky (Pelkki and Arthaud 1998; Luppold and Baumgras 1999). Thus, expansion of the beaver population in Kentucky could have severe consequences for the timber industry of the state if beavers cut and girdle valuable timber species. In this paper we present data on the impact on woody plant species of two recently established beaver colonies at Berea Forest in eastern Kentucky.

### STUDY AREA AND METHODS

Berea Forest is located on the western edge of the Cumberland Plateau in Jackson and Madison counties, Kentucky. The forest is owned by Berea College and includes the Owsley Fork Reservoir. The reservoir, 61 ha in size and 12 m at maximum depth, was constructed in the early 1970s and now serves as the primary water supply for the city of Berea, Kentucky; beavers first moved into the reservoir in the mid-1990s (J. Perry, Berea College, pers. comm.), and have established colony sites at two locations: a bank den at the edge of Radford Hollow and a lodge at the confluence of Owsley Fork and the reservoir. Berea Forest is situated in the mixed mesophytic forest region (Braun 1950), and supports second-growth hardwood forests about 30–45 yrs in age. Dominant tree species include yellow-poplar, white oak (*Quercus alba*), and sugar maple. American beech, white ash (*Fraxinus americana*), Virginia pine (*Pinus virginiana*), chestnut oak (*Q. prinus*), northern red oak (*Q. rubra*), and black oak (*Q. velutina*) also are common.

From 18 January to 15 February 2000 we sampled woody plant species for the presence of beaver cutting, sampling, and feeding activity. An initial reconnaissance identified the location of colony sites and the limits to which beaver activity was observed from each colony site and from the shoreline. We established a 10% area cruise along the shore of the reservoir and sampled twenty-five 0.04-ha plots placed tangentially to the shoreline in the zone

where feeding activity by beavers took place. Distance between plots was 177 m. Plots were divided into proximal and distal halves to assess changes in feeding patterns of beavers with increasing distance from the shore. Within plots, all stems were recorded by species, diameter at 1 m above ground, distance from shore, and extent of damage. Diameter of stumps was measured at the top of the stump. Damage categories assigned to stems were uncut (no damage visible), barked (damage to the bark or xylem), and cut (completely felled or felled and removed leaving the stump behind). Within the barked category, no distinction was made between stems that represented food sampling and those that reflected food consumption. Damaged stems were assigned to size classes as follows:  $\leq 4$  cm dbh;  $>4$  to  $\leq 14$  cm dbh;  $>14$  to  $\leq 24$  cm dbh; and  $>24$  cm dbh. Using a Chi-square test, we compared the frequency of selection of woody plant species with the availability of stems. We used a Bonferroni z-statistic to determine if the frequency of selection among plant species occurred more or less than expected (Neu et al. 1974). A test was considered significant when  $P < 0.05$ .

### RESULTS AND DISCUSSION

Beavers damaged 18 species of woody plants, with Virginia pine ( $n = 42$ ), willow ( $n = 33$ ), and American beech ( $n = 29$ ) affected most frequently (Table 1). Beavers exhibited selectivity in plant choice ( $\chi^2 = 14.9$ ,  $df = 6$ ,  $P < 0.025$ ), with yellow-poplar and oaks avoided (Table 2). All other plant species or species groups were damaged at proportions consistent with their availability in the habitat. Of the 173 stems showing evidence of damage, 94.8% were located in the proximal half of sample plots, indicating that beavers restricted much of their feeding activity closer to the shoreline. Of the stems showing evidence of damage, 19% ( $n = 33$ ) were barked and 81% ( $n = 140$ ) were cut. Of the cut stems, 97% ( $n = 136$ ) were removed from the site. Beaver fed most frequently on stems  $<4$  cm dbh ( $n = 105$ ) and  $>4$  to 14 cm dbh ( $n = 48$ ), and less frequently on stems  $>14$  to 24 cm dbh ( $n = 17$ ) and  $>24$  cm dbh ( $n = 3$ ). The largest stem showing evidence of beaver feeding activity was an American beech at 36 cm in diameter.

Table 1. Availability of woody plant species (*n* = 284) in relation to plant species damaged (*n* = 173) by beaver in Berea Forest, Jackson County, Kentucky, 2000.

Species	Number available (% of total available)	Number consumed (% of total consumed)
<i>Acer rubrum</i>	10 (3.5)	5 (2.9)
<i>A. saccharum</i>	13 (4.6)	8 (4.6)
<i>Acer</i> spp.	6 (2.1)	6 (3.5)
<i>Aesculus octandra</i>	1 (0.4)	1 (0.6)
<i>Betula lenta</i>	3 (1.0)	2 (1.1)
<i>Carpinus caroliniana</i>	4 (1.4)	4 (2.3)
<i>Carya</i> spp.	3 (1.0)	2 (1.1)
<i>Cornus florida</i>	4 (1.4)	3 (1.7)
<i>Fagus grandifolia</i>	42 (14.9)	29 (16.8)
<i>Fraxinus americana</i>	9 (3.2)	4 (2.3)
<i>Juniperus virginiana</i>	2 (0.7)	0 (0.0)
<i>Liriodendron tulipifera</i>	22 (7.7)	6 (3.5)
<i>Ostrya virginiana</i>	1 (0.4)	1 (0.6)
<i>Oxydendrum arboreum</i>	1 (0.4)	0 (0.0)
<i>Pinus strobus</i>	1 (0.4)	1 (0.6)
<i>P. virginiana</i>	67 (23.6)	42 (24.3)
<i>Platanus occidentalis</i>	10 (3.5)	3 (1.7)
<i>Quercus alba</i>	13 (4.6)	3 (1.7)
<i>Q. coccinea</i>	5 (1.8)	0 (0.0)
<i>Q. palustris</i>	2 (0.7)	2 (1.1)
<i>Q. prinus</i>	1 (0.4)	0 (0.0)
<i>Q. rubra</i>	1 (0.4)	0 (0.0)
<i>Robinia pseudoacacia</i>	2 (0.7)	0 (0.0)
<i>Salix</i> spp.	37 (13.0)	33 (19.1)
<i>Ulmus</i> spp.	11 (3.9)	5 (2.9)
Unidentified	13 (4.6)	13 (7.5)

Beavers avoid or feed little on coniferous plant species in many portions of their range (Belovsky 1984; Brenner 1962; Hall 1960; Jenkins 1980; Northcott 1971; Shiples et al. 1979); however, consumption of conifers in spring is known to occur (Jenkins 1979; Kienzler 1971; Nixon and Ely 1969; Roberts and Arner 1984; Svendsen 1980). Typically, beavers debark pines either to feed on the cambium (Nixon

and Ely 1969) or to lick off the exposed pitch after the bark is removed (Svendsen 1980). Jenkins (1979) suggested that the value of bark of coniferous trees is likely higher than for deciduous trees in spring, as coniferous species build up carbohydrate reserves in winter whereas deciduous species exhibit a sharp drop in carbohydrate reserves in spring (Koslowski and Keller 1966; Kramer and Koslowski 1960). Although Virginia pine was not damaged by beavers relative to its availability, we found Virginia pine to be the dominant woody plant species impacted by beavers with many of the stems cut and removed. In Louisiana, Chabreck (1958) also found a coniferous species, loblolly pine (*Pinus taeda*), to compose the largest proportion of the diet of beavers.

Evidence suggests that beavers exhibit spatial feeding patterns consistent with optimal foraging theory (Schoener 1979), with the pattern reversed in beavers such that larger stems are selected closer to the den site and smaller stems at more distant locations (Belovsky 1984; Fryxell and Doucet 1993; Jenkins 1980; Pinkowski 1983). Our analysis could not readily confirm this, but we observed a strong selection by beavers for smaller-diameter stems over larger-diameter stems, especially close to the shoreline. In fact, most feeding activity was concentrated within 4 m of the shore, regardless of how far a segment of shore was from the den site. A similar pattern in feeding behavior of beaver was reported by Shiples et al. (1979).

Harvest and processing of commercial timber continues to be an important economic industry in Kentucky, particularly in the eastern Cumberland Plateau region (Luppold and

Table 2. Confidence intervals and outcomes of selection tests of woody plants damaged by beavers in Berea Forest, Jackson County, Kentucky, 2000.

Species/Species group <sup>1</sup>	Proportion of available habitat	Confidence interval	Cutting behavior
<i>Acer</i> spp.	0.102	0.051 ≤ <i>p</i> ≤ 0.169	Random
<i>Fagus grandifolia</i>	0.149	0.099 ≤ <i>p</i> ≤ 0.236	Random
<i>Liriodendron tulipifera</i>	0.077	0.001 ≤ <i>p</i> ≤ 0.069	Avoided
<i>Pinus virginiana</i>	0.236	0.162 ≤ <i>p</i> ≤ 0.324	Random
<i>Quercus</i> spp.	0.078	-0.003 ≤ <i>p</i> ≤ 0.061	Avoided
<i>Salix</i> spp.	0.130	0.117 ≤ <i>p</i> ≤ 0.263	Random
Other	0.229	0.147 ≤ <i>p</i> ≤ 0.303	Random

<sup>1</sup>Species groupings included all maples combined (*Acer* spp.), all oaks combined (*Quercus* spp.), and other (all species not observed in sufficient number for analysis separately)

Baumgras 1999; Pelkki and Arthaud 1998). Despite the consumption of valued timber species by beavers, including yellow-poplar, in adjacent states (Nixon and Ely 1969; Svendsen 1980), we observed little damage by beavers on commercially valuable timber species. Beavers avoided yellow-poplar and oaks and extensively damaged small-diameter stems, suggesting that beavers are having minimal effect on the standing timber resource in Berea Forest. An explanation for the avoidance of yellow-poplar remains unclear. Berea College harvests timber on an irregular basis from Berea Forest as a means of providing additional support funds for the college (J. Perry, Berea College, pers. comm.). Studies on beavers in other locations in eastern Kentucky are now needed to confirm what the long-range effects on timber resources will be from expanding beaver populations in the state.

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## Genetic Polymorphism in the Mitochondrial Gene Cytochrome b in Kentucky *Peromyscus* (Deer Mouse) Populations

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### ABSTRACT

Three species of *Peromyscus* are known in Kentucky: *P. maniculatus*, *P. leucopus*, and *P. gossypinus*. The current method of distinguishing them is through the use of morphological characteristics. In this study, a genetic method of distinguishing between morphologically similar species of *Peromyscus* was developed based on percent sequence identity of the cytochrome b gene. Several species are known to hybridize in the wild and in the laboratory. *Peromyscus leucopus* specimens were trapped at the Blue Grass Army Depot in Madison County, KY; *P. gossypinus*, in Henderson County, KY. PCR was used to amplify the gene cytochrome b using DNA isolated from wild-caught specimens and from laboratory stocks. PCR was followed by restriction enzyme digestion with *AseI*. PCR products were sequenced and NCBI BLAST sequence alignment analysis was performed to determine percent identity with sequences in GenBank and between the native specimens. Wild caught *P. leucopus* showed 98% identity with the *P. gossypinus* sequence in GenBank; native *P. gossypinus* showed 94% identity for cytochrome b. Comparisons made with *P. maniculatus* showed 89% identity. Results show that Kentucky specimens are genetically distinguishable based on genetic polymorphism.

### INTRODUCTION

The genus *Peromyscus*, native to North America, has ca. 60 species occurring in almost all parts of the United States (Hall 1981). Three species of *Peromyscus* exist throughout Kentucky, thriving in various geographical locations. Of the three, *P. maniculatus*, deer mouse, and *P. leucopus*, white-footed mouse, are known to hybridize in nature (Hall 1981). Another Kentucky species, *P. gossypinus*, cotton mouse, also hybridizes with *P. leucopus* (Dawson et al. 1993). One problem with studying *Peromyscus* in Kentucky is being able to distinguish one species or subspecies from another. Such is the case with *P. m. nubiterrae* and *P. l. leucopus*, it being difficult to distinguish the two species solely on morphological methods. But they should be readily distinguishable using their DNA (Barbour and Davis 1974). *Peromyscus maniculatus* populations are abundant in open weedy fields, grasslands, and agricultural land throughout most of Kentucky. They are known to occur in the western and central regions, but are report-

edly absent from the southeastern mountains and adjacent parts of the Cumberland Plateau. Two subspecies of *P. maniculatus* are known in Kentucky, *P. m. bairdii* and *P. m. nubiterrae*. They are separated geographically but can be difficult to distinguish based on morphology (Barbour and Davis 1974). *Peromyscus m. nubiterrae* has been documented only in the Big Black Mountain region of Harlan County; *P. m. bairdii* can be found in central Kentucky (Barbour and Davis 1974).

Of the subspecies of *P. leucopus* in Kentucky, *P. l. leucopus* occurs in the western half of the state; *P. l. noveboracensis*, in the eastern half. These subspecies appear to intergrade in the Bluegrass region (Barbour and Davis 1974). *Peromyscus m. nubiterrae* is often confused with *P. l. leucopus* in areas where the species occur sympatrically. Ideally, the two species should be distinguishable based on body size (*P. leucopus* is larger), but occasionally individuals are difficult to distinguish based on morphology (Rich et al. 1996).

In western Kentucky there is a third species of *Peromyscus*, *P. gossypinus*, cotton mouse,

CAATGATTCCTTCATTGATCTCCCAACCCCATCCAATATCTCATCATGATGAAACT  
 TCGGATCCTTACTTGGACTGTGCCTAGTAATTCAAATTTAACTGGCCTATTCTTAGCCATACAC  
 TACACATCCGACACAACACTACAGCATTCTCATCCGTAACACATATCTGCCGAGACGTAAACTACGG  
 ATGACTAATCCGATATATACACGAAACGGAGCCTCAATATTC  
 TTTATCTGCTTATTCTGCACGTA  
 CAATGATTCCTTCATTGATCTCCCAACCCCATCCAATATCTCATCATGATGAAACTTCGGATCCT  
 TACTTGGACTGTGCCTAGTAATTCAAATTTAACTGGCCTATTCTTAGCCATACACTACACATCC  
 GACACAACACTACAGCATTCTCATCCGTAACACATATCTGCCGAGACGTAAACTACGGATGACTAAT  
 CCGATATATACACGCAACGGAGCCTCAATATTCTTTATCTGCTT

Figure 1a. GenBank Sequence for cytochrome b gene in *P. leucopus* (253 bases). Bold letters indicate *ApoI* restriction site starting at position 90, which should produce two fragments, one 91bp and the other 162bp in length. *gi|9274771embIX89790IMTPLYB* *P. leucopus* mitochondrial DNA for cyt b gene Length = 321.

Figure 1b. GenBank Sequence for cytochrome b gene in *P. gossypinus* (240bp). There is no restriction site, producing only one larger fragment. *embIX89786.1IMTPGCTYB* *P. gossypinus* mitochondrial DNA for cytochrome b gene. Length = 321.

which occurs in lowlands, wooded stream banks, swampy woods, and brushland of the Purchase Region. The habitat of *P. gossypinus* overlaps that of *P. leucopus* (Barbour and Davis 1974). Although the two mice are clearly separate species, it has been observed that they can hybridize in the lab (Foster 1995), and natural hybrids from Dismal Swamp were reported by Dice (Hall 1981).

The objective of our project was to analyze the genetic diversity of native *Peromyscus* populations in Kentucky by detecting polymorphisms that could then be used as genetic markers (Arnheim et al. 1990).

#### MATERIALS AND METHODS

DNA was isolated from ca. 66 *Peromyscus* specimens recovered in Kentucky between October 1996 and May 1997 from study sites including the Blue Grass Army Depot (Madison County); Maywoods Environmental Laboratory, Morehead (Rowan County); Cave Run Lake; Campbell County; and Henderson County. Using standard field trapping techniques, we collected native deer mice in Sherman live traps. As a control, stock specimens were obtained from the *Peromyscus* Genetic Stock Center at the University of South Carolina. A 2.54 cm clipping from the distal end

of each specimen's tail was used as a source of DNA by following procedures modified from Sambrook (1989); the concentration of each DNA sample was determined using a spectrophotometer. PCR amplifications were carried out in a Perkin Elmer 2400 Thermocycler using a procedure modified from Sambrook (1989). The amplified DNA was then run on an agarose gel and visualized with ethidium bromide.

Custom primers were designed to amplify the mitochondrial gene cytochrome b in *Peromyscus* based on sequence data obtained from GenBank (Sullivan et al. 1995). The cytochrome b gene in *P. gossypinus* contains an *ApoI* restriction site (Figures 1a, 1b). Successful digestion with *ApoI* produces two visible bands. There is no *ApoI* restriction site in the cytochrome b sequence for *P. leucopus*, resulting in a single, larger band visible on a 2% agarose gel.

Following PCR amplification, agarose gel electrophoresis was conducted to determine whether or not an appropriate sized product was obtained using a primer pair consisting of sense and anti-sense primer sequences. On a 12 × 15 cm agarose gel, a band could be considered distinct from another band if and only if the other band was 1 mm or greater in dis-

tance above or below the position of the original band (Westneat 1990). Band sizes were estimated using a 100 bp DNA ladder (Promega). After enzyme digestion, a portion of the sample was again run on a 2% agarose gel, using 1× TBE buffer, at ca. 85 volts for 2 hours to view any banding PCR-RFLP patterns (Sambrook et al. 1989).

Genetic similarity (i.e., the extent to which nucleotide sequences are related) between two morphologically similar species of *Peromyscus* was assessed based on percent sequence identity (the extent to which the two nucleotide sequences are invariant). PCR products from *P. gossypinus* and *P. leucopus* were sequenced using an ABI PRISM system. Once the nucleotide sequences were known, NCBI sequence alignment analysis was performed to determine percent identity with sequences in the GenBank database.

## RESULTS

DNA from the *Peromyscus* specimens was isolated and concentration was determined before PCR amplification and restriction enzyme digestion. There was successful amplification of the cytochrome b gene in all of the *Peromyscus* specimens. There is obvious polymorphism in nucleotide sequences between native Kentucky *Peromyscus* species (Figures 2a–2d) and between Kentucky species and those represented in GenBank (Figures 1a, 1b). The sequences from both Kentucky native specimens contain the *ApoI* restriction site at position 90 in the upper strands and also in similar but distinguishable positions in the lower strands, making native Kentucky specimens distinguishable based on this polymorphism.

Through the use of BLAST, wild caught *P. leucopus* showed 98% identity with the *P. gossypinus* sequence in GenBank; native *P. gossypinus* showed 94% identity. Comparisons made with *P. maniculatus* showed 89% identity; with *P. polionotus* (obtained from the Stock Center), 91% sequence identity.

## DISCUSSION

PCR-RFLP using custom primers designed to amplify the cytochrome b gene in horses (a 590 base pair fragment) has been successfully applied to analysis of thoroughbred horses. This has made it possible to classify thorough-

bred horses into four types (Ishida et al. 1996) based on genetic polymorphism. This gene is highly conserved as expected, and in the nucleotide sequence of native Kentucky specimens there is genetic polymorphism, which may be used as a genetic marker. The *P. leucopus* sequence from GenBank has the *ApoI* restriction site while the *P. gossypinus* from GenBank does not, making this a potentially useful marker for distinguishing the species. There is obvious polymorphism in nucleotide sequences between native Kentucky *Peromyscus* species (Figures 2a–2d), and between Kentucky species and those represented in GenBank (Figures 1a, 1b). The sequences from both Kentucky native specimens contain the *ApoI* restriction site at position 90 in the upper strands and also in similar but distinguishable positions in the lower strands, making native Kentucky specimens distinguishable based on this polymorphism.

## SUMMARY

The development of species specific genetic markers like the ones attempted in this study can have numerous useful applications in population biology. Examples include (1) improved resolution of species identification when combined with morphological approaches and (2) improved species conservation efforts by enhancing detection of genetic differences among populations.

As for the custom primers, the mitochondrial gene cytochrome b was selected after an extensive GenBank search for potentially suitable markers. Although mitochondrial genes are not useful in diagnosing interspecific hybridization per se (because mitochondrial DNA is primarily maternally derived), when used in combination with genomic markers, the DNA can present a more complete picture of hybridization because it can indicate the direction of the interspecific crosses. The PCR data produced in this study showed no contamination and produced a discreet band that was of the expected size, thus indicating proper amplification of the cytochrome b gene.

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CAATGAACCTTCATTGATCTCCCAACCCCATCCAATATCTCATCATGATAATCGGATCCTTAC  
 TTGGACTGTGCCTAGTAATTC**AAATT**TAACTGGCCTATTCTTAGCCATACACTACACATCCGAC  
 ACAACTACAGCATTCTCATCCGTAACACATATCTGCCGAGACGTAAACTACGGATGACTAATCCG  
 ATATATACACGCAAACGGAGCCTCAATATTCTTTATCTGCTTATTCTGCACGTANGACGAGGAA  
 TATACTACGGATNCTANCNNTNCAGG AATANNCANATN  
 TCCTACGTGCAGGAATAAGCAGATAAAGAATATTGAGGCTCCGTTTGCCTGTATATATCGGGATT  
 AGTCATCCGTAGTTTACGTCTCGGCAGATATGTGTTACGGATGAGAATGCTGTAGTTGTGTCGGA  
 TGTGTAGTGTATGGCTAAGAATAGGCCAGTT**AAATT**TGAATTACTAGGCACAGTCCAAGTAAGG  
 ATCCGAAGTTTCATCATGATGAGATATTGGATGGGGTTGGGAGATCAATGAAGGATTCATTGATA  
 ATTTTAAGTAGTGGGTGTANNNNNNNNN  
 GTCCTACGTGCAGGAATAAGCAGATAAAGAATATTGAGGCTCCGTTTGCCTGTATATATCGGGATT  
 AGTCATCCGTAGTTTACGTCTCGGCAGATATGTGTTACGGATGAGAATGTGTAGTTGTGTCGGAT  
 GTGTAGTGTATGGCTAAGAATAGGCCAGTT**AAATT**TGAATTACTAGGCACAGTCCAAGTAAGGA  
 TCCGAAGTTTCATCATGATGAGATATTGGATGGGGTTGGGAGATCAATGAAGGATTCATTGATAA  
 TTTTAAGTAGTGGGTGANNNNNNNNNNNNNNNNNNNNNNN  
 CAATGATTCTTCATTGATCTCCCAACCCCATCCAATATCTCATCATGATAAACTTCGGATCCT  
 TACTTGACTGTGCCTAGTAATTC**AAATT**TAACTGGCCTATTCTTAGCCATACACTACACATCC  
 GACACAACACTACAGCATTCTCATCCGTAACACATATCTGCCGAGACGTAAACTACGGATGACTAAT  
 CCGATATATACACGCAAACGGAGCCTCAATATTCTTTATCTGCTTATTCTGCACGTANGACGAG  
 GAATATACTACGGGATNCTACGTNNANGNNTANCNAANN

Figure 2a. 300 bases of the cytochrome b DNA sequence for *Peromyscus leucopus* from the Blue Grass Army Depot in Madison County, KY. Bold letters indicate *ApoI* restriction site starting at position 89 (#38 U). N stands for any base.

Figure 2b. 300 bases of the cytochrome b DNA sequence for *Peromyscus leucopus* from the Blue Grass Army Depot in Madison County, KY (#38 L). Bold letters indicate *ApoI* restriction site starting at position 142 creating two fragments, one 143bp and the other 157bp in length. N stands for any base.

Figure 2c. 300 bases of the cytochrome b DNA sequence for *Peromyscus gossypinus* (#62 L) from Henderson County, KY. Bold letters indicate *ApoI* restriction site starting at position 165, creating two fragments, one 166bp and the other 134bp in length. N stands for any base.

Figure 2d. 300 bases of the cytochrome b DNA sequence for *Peromyscus gossypinus* (#62 U) from Henderson County, KY. Bold letters indicate *ApoI* restriction site starting at position 89, creating two fragments, one 90bp and the other 134bp in length. N stands for any base.

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# Empirical Measurements of the Antenna Gain of the Morehead Radio Telescope

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## ABSTRACT

The Morehead Radio Telescope (MRT) is a filled-aperture radio telescope designed to support undergraduate research. The instrument consists of a  $13.25 \times 3.35$  meter parabolic antenna, an automated alt-azimuth positioning system, a state-of-the-art front and back-end hydrogen-line receiver system (operating at 1420 MHz), and supporting electronics and hardware. The sensitivity and versatility of the telescope design facilitate the investigation of a wide variety of astrophysically interesting phenomena. The performance characteristics of the telescope have now been empirically measured and allow a varied and in-depth scientific program. The power gain of a telescope is, in part, a measure of the directivity of the antenna and how the telescope converts radiation energy to output energy. Measurements of antenna gain are critical to understanding important telescope characteristics including the instrument's overall sensitivity, which is measured by the minimum detectable flux density. A series of experiments was performed to measure the MRT antenna gain by a variation of the standard three-antenna method. A dipole antenna of known gain was compared to a calibration horn antenna constructed for these experiments which was, in turn, compared to the MRT antenna. Results from these experiments have been completed and are herein reported.

## INTRODUCTION

The intent of the Morehead Radio Telescope (MRT), Morehead State University (MSU), Morehead, Kentucky, is to provide an instrument capable of supporting scientific research in observational astrophysics at radio frequencies. The telescope has been in operation since 1997. Among the most important characteristics that determine a radio telescope's performance are the antenna's radiation (beam) pattern and antenna gain. The radiation pattern is a three-dimensional representation of the magnitude, phase, and polarization of the antenna's directivity function and therefore determines the instrument's spatial resolution, which is typically expressed as the half power beamwidth (HPBW), that is, the width of the sensitivity profile at half maximum. Empirical measurements of the radiation pattern have been previously reported

(Malphrus et al. 1999). The antenna gain, measured in decibels, is a measure of how the telescope amplifies received. The intent of this experiment was to complete a series of experiments to empirically measure the MRT gain and determine the instrument's sensitivity as a function of the empirical antenna gain. These experiments were based upon the standard 3-antenna method of antenna measurement in which the gain of an antenna that is unknown is measured through a series of "bootstrap" tests that compare antennas to a standard antenna whose gain is well understood. In this series of experiments, a dipole antenna of known gain was compared to a horn (calibration) antenna constructed for these experiments, which was, in turn, compared to the MRT antenna. Relative power gains between antennas could then be established. As the power gain of the dipole antenna is well un-



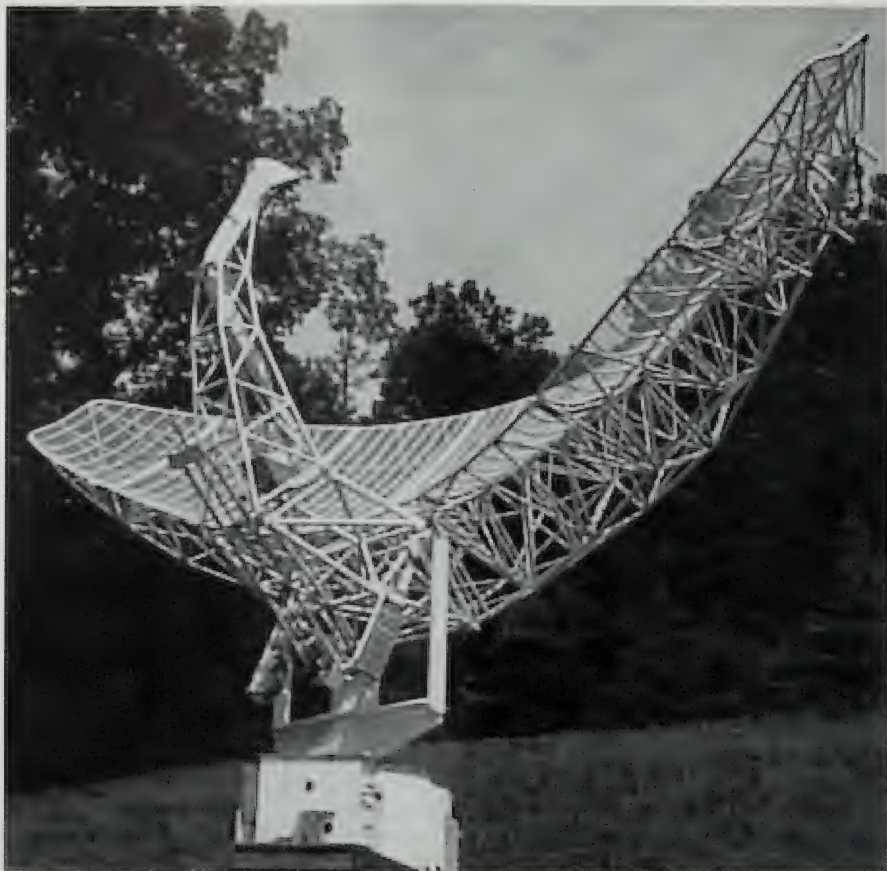


Figure 1. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky.

derstood, an absolute gain for the MRT antenna could then be measured.

#### MRT INSTRUMENTATION

The MRT system includes a wire-mesh parabolic reflecting antenna (Figure 1), alt-azimuth tracking positioner control and drive systems, receiver and signal processing system, a controlling computer and data acquisition (DAQ) system, and supporting electronics and hardware. The current observing scheme was a total power receiver that converts radiation from space concentrated by the antenna system to an electrical signal, that is amplified,

modified, and interpreted. The MRT system is controlled by a 400 MHz Pentium-based controlling computer that uses a National Instruments PC1-M10-16E-4 DAQ interface board, optical isolation system, and robotic drive and control systems developed by MSU faculty and students. The control system computer points the telescope, tracks the cosmic sources, and controls data collection and storage. The data from a particular experiment is then written in a spreadsheet format on a 3-D modeling program HI-Q, which is then used to produce contour and topographic images of the HI distribution of cosmic sources.

The telescope uses a total power receiver system operating at 1420 MHz with a 6 MHz bandwidth. The measured and estimated performance characteristics of the system are given: spatial resolution =  $1.2 \times 3.6$  arcdegrees (Malphrus *et al.* 1998), total receiver system temperature = 67.3 K (Kruth 1998), and the theoretical minimum detectable flux density = 4.3 Jy. The instrument has been described in detail in an earlier publication (Malphrus *et al.* 1998). The current experiment utilized the MRT system, a dipole antenna, a horn antenna constructed on site, and a remotely operated 1420 MHz radio frequency transmitter. The primary components of the system used are summarized below.

#### Antenna

The MRT employs a high-gain, 40-foot antenna initially designed for L-Band operation. An Army NIKE-Hercules ANS-17 Radar antenna was modified for radio astronomy applications. The basic system includes a parabolic reflector, antenna feed horn, waveguide system, and azimuth-elevation positioning system. The current, third-iteration-positioning system provides azimuth coverage of  $360^\circ$  at a maximum antenna rotation speed of one revolution in 12 minutes and elevation coverage of  $0$  to  $90^\circ$  in 10 minutes. The positioning system also allows for continuous drive in tracking mode, essentially  $1^\circ$  in 4 minutes at the celestial equator and slower by the cosine of the declination as the angle increases or decreases.

#### Remote 1420 MHz Transmitter

A 1420 MHz transmitter was designed and constructed to generate an artificial signal to sweep the antenna beam through to determine the directivity pattern. The same transmitter was initially used for the current experiments. The transmitter was designed and constructed by K-MEC and Morehead State University students. The device uses a voltage oscillator to create the 1420 MHz signal. The voltage oscillator circuit requires a 12-V power supply, a voltage regulator and a 317-tuning regulator. A 12-V battery is used to power a DC/DC converter. The DC/DC converter has an output of +9 V and -9 V. The -9 V is looped back around into the converter to double the negative voltage output. The main ob-

stacle posed by the use of the voltage regulator was its thermal instability. This arrangement causes a problem because the 12-V battery and the DC/DC converter both produce heat, which varies the output of the voltage regulator. The 317 tuning regulator is utilized to overcome this limitation. It can add from 0 to 18 V depending on what is required to keep the transmitter producing the correct frequency. To further lessen the thermal effects on the transmitter, it was placed in a thermally stable housing with Styrofoam surrounding it to absorb any heat from the environment. The final iteration produced a phase stable radiation source at 1420 MHz that allowed the radiation pattern and an initial series of gain measurement experiments to be performed.

The 1420 MHz transmitter was initially used to compare the gain of a dipole antenna to the gain of the calibration (horn) antenna. The 1420 MHz transmitter, however, provided insufficient power to complete phase two of the experiment (the comparison of the horn antenna to the MRT). The transmitter was required to provide effective detectable radiant power even when located at the distance dictated by the MRT antenna's far field pattern (discussed later in detail). The 1420 MHz transmitter failed to generate sufficient power at the great distance required to make the measurement with a high level of precision. A commercial HP 8164A RF signal generator proved to provide sufficient power levels and phase stability to complete the experiment. Phase one was repeated using the commercial transmitter.

#### THEORETICAL PERFORMANCE CHARACTERISTICS OF THE MRT ANTENNA

The response of an antenna as a function of direction is described by its radiation pattern. This pattern is the same for receiving as it is for transmitting and is known as antenna reciprocity. Typically, there is one main lobe and several smaller lobes. The evaluation of the radiation pattern and lobes it contains can reveal important characteristics of the antenna's performance. To measure this pattern, a transmitter must be used to create the desired frequency. There is a point in the measurement process where the pattern does not change if the transmitter is moved any farther away.



This response pattern is called the Fraunhofer or far-field pattern. Inside this distance, the near field pattern (which varies as a function of angle and distance), is measured. To successfully complete the measurement requires that the transmitter be placed sufficiently far away so that the Fraunhofer pattern is observed. This minimum range is given by equation (1):

$$S_{\min} = \frac{2D^2}{\lambda} \quad (1)$$

where  $S_{\min}$  is the minimum range distance,  $D$  is the largest aperture dimension of the antenna, and  $\lambda$  is the wavelength (Kraus 1986). Using 13.25 m for  $D$  and 0.2112 m for the wavelength, the minimum range distance is 1662 m (The ARRL Antenna Book 1994). Empirical measurements of the radiation pattern of the MRT have been previously reported (Malphrus et al. 1998).

Perhaps the most important characteristic of an antenna's properties is the quantity of energy it concentrates preferentially in one direction. This property, called an antenna directivity, is equal to its power gain if the antenna is 100% efficient. Power gain is often expressed relative to a reference value such as an isotropic radiator of a half-wavelength dipole. The power radiated by an antenna is given by the general formula

$$P_r = \frac{1}{2} \text{Re} \iint (E \times H^*) ds \quad (2)$$

where  $P_r$  is the radiated power,  $\text{Re}$  is the reflectivity constant for the antenna,  $E$  is the electric field,  $H$  is the magnetic field, and  $ds$  is the line integral. Translated into spherical coordinates, which are more relevant to real world problems, the relationship becomes

$$P_r = \frac{1}{2} \text{Re} \iint (E_\theta H_\phi^* - E_\phi H_\theta^*) r^2 \sin \theta d\theta d\phi. \quad (3)$$

The directive gain, which is directly related to the power gain, is defined as the ratio of the radiation intensity in a certain direction to the average radiation intensity (Stutzman 1981). Directive gain is defined by the relationship

$$D(\theta, \phi) = \frac{U(\theta, \phi)}{U_{\text{ave}}} \quad (4)$$

$D(\theta, \phi)$  is the directive gain as a function angle,  $U(\theta, \phi)$  is the maximum radiation intensity (in watts  $\text{sr}^{-1}$ ), and  $U_{\text{ave}}$  is the average radiation intensity (also in watts  $\text{sr}^{-1}$ ) (Kraus 1986). In general, equation (4) shows that the radiation pattern has both a  $\theta$  and a  $\phi$  component. If both the numerator and the denominator are divided by  $r^2$ , equation (4) gives power densities. The directive gain is now the ratio of the power density in a particular direction at a specific range,  $r$ , to the average power density at the given range  $r$  (Stutzman 1981)

$$D(\theta, \phi) = \frac{U(\theta, \phi)/r^2}{U_{\text{ave}}/r^2};$$

$$D(\theta, \phi) = \frac{\frac{1}{2} \text{Re}(E \times H^*) \cdot r}{P_r/4\pi r^2}. \quad (5)$$

Through some basic substitutions, the antenna-beam solid angle,  $\Omega_A$ , can be found.  $\Omega_A$  is the solid angle through all the power would be radiated if the power per unit solid angle (radiation intensity) equaled the maximum value over the beam area. The directivity is now defined as maximum value of the directive gain. By following the substitution below the directivity can be expressed in terms of  $\Omega_A$  (Kraus 1986)

$$D = \frac{U_{\text{in}}}{U_{\text{ave}}},$$

substituting  $U_{\text{ave}} = P_r/4\pi$

$$D = \frac{U_{\text{m}}}{P_r/4\pi} = \frac{4\pi U_{\text{m}}}{U_{\text{m}} \Omega_A},$$

reducing yields

$$D = \frac{4\pi}{\Omega_A}. \quad (6)$$

When an antenna is used for transmitting or receiving a signal, how efficiently the antenna transforms the available power at its input to radiated power is defined as the gain. Gain is defined as  $4\pi$  times the ratio of the radiation intensity in a specified direction to the net power accepted by the antenna from a connected transmitter and is described by equation (7)

$$G(\theta, \phi) = \frac{4\pi U(\theta, \phi)}{P_{\text{in}}}. \quad (7)$$

$G(\theta, \phi)$  is the gain,  $P_{in}$  is the input power accepted by the antenna, and  $U(\theta, \phi)$  is the radiation intensity of the antenna in a specific direction  $(\theta, \phi)$ . Using the definition of directivity and antenna beam solid angle, the maximum value of the power gain can be attained by

$$G = \frac{4\pi U_m}{P_{in}}. \quad (8)$$

The only difference between power gain and directivity is the power value used. The power gain depends on  $P_{in}$ , the input power, while the directivity depends on the  $P_r$ , the radiated power. If all the power received by the antenna at the inputs were radiated, then  $P_{in}$  would equal  $P_r$ . The power gain takes into account the fact that real antennas lose some of the input power before it can be radiated. The fraction of input power lost is absorbed by the antenna and nearby structures. Because of this loss, the power gain and the directivity can be related by  $e$ , the radiation efficiency

$$e = \frac{P_r}{P_{in}}. \quad (9)$$

Using the above equations, the power gain and the directivity can be related (Stutzman 1981),

$$G = eD. \quad (10)$$

### Gain Measurements

The power gain measurement of an antenna is an absolute measurement (verses a relative measurement) which makes it a more difficult characteristic to attain. One method is to take a source of constant power and let it drive a source antenna. Next, an antenna of known gain is attached to a spectrum analyzer and a power measurement is taken. Then, the antenna with unknown gain is then attached to the spectrum analyzer and another power measurement is taken. The difference on the spectrum analyzer between the two measurements is the relative difference in gain (dB) between the two antennas. Finally, the gain of the unknown antenna is then found by adding the difference between the two to the gain of the known antenna. The three-antenna method extends this experiment by bootstrapping the measurements to determine the MRT antenna gain.

### Horn Antenna Design

To obtain the gain of the Morehead Radio Telescope, a horn antenna of known gain had to be designed for a reference gain. A pyramidal horn antenna ( $E$  and  $H$  plane) was designed based on the geometry in Figure 2. A gain of 20 dB was chosen because any larger gain would yield an antenna that was too large to manage. Using the equation below, the dimensions of the horn antenna can be found (Stutzman 1981)

$$\begin{aligned} \left[ \sqrt{2\sigma} - \frac{b}{\lambda} \right]^2 (2\sigma - 1) &= \left( \frac{G}{2\sqrt{2\pi}} \frac{1}{\sqrt{\sigma}} - \frac{a}{\lambda} \right)^2 \\ &\times \left( \frac{G^2}{18\pi^2} \frac{1}{\sigma} - 1 \right) \\ A &= \sqrt{3M_H} \\ B &= \sqrt{2M_E} \\ G &= \frac{1}{2} \frac{4\pi}{\lambda^2} (AB). \end{aligned} \quad (11)$$

Using the known values  $G = 200$  and the dimensions of the waveguide,  $b = 0.083312$  m and  $a = 0.165$  m, other dimensions included:  $B = 0.74425$  m,  $A = 0.953868$  m,  $l_E = 1.3113$  4 m, and  $l_H = 1.436$  m. 16 gauge aluminum was utilized in the construction of the reflecting horn. A support structure in the shape of the pyramidal horn antenna was first constructed of 0.5" plywood. The aluminum reflecting surface was bonded to the structural surface with a multi-purpose adhesive. The pyramidal structure was further supported by a 2"  $\times$  4" wooden frame which also aided in its transportation to the antenna test range.

### Dipole Antenna Design

Although the horn antenna was designed for a gain of 20 dB, it needed to be experimentally verified before being used to measure the gain of the MRT. A dipole antenna was designed for this purpose. The classic method of using a quarter wave brass elements and co-axial hardline was employed in the construction of the dipole antenna. The antenna was tuned to length by using a Hewlett Packard 8614A signal generator, a spectrum analyzer, and a Hewlett Packard 5245L electronic frequency counter. The antenna was coupled to the signal generator and the spectrum analyzer by a



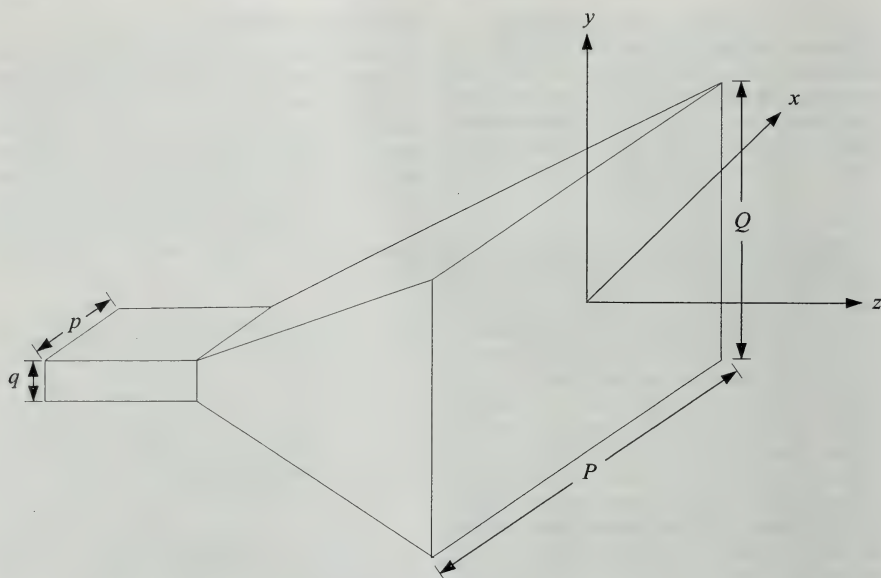


Figure 2. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. Schematic drawing of the calibration horn antenna, with dimensions  $P = 0.9539$  m,  $Q = 0.7443$  m,  $q = 0.08331$  m, and  $p = 0.165$  m.

RF-coupling. The directivity of the coupler was measured to be 30 dB at 1.4 GHz. The transmitted signal was tuned by shortening the brass elements until the desired frequency was obtained.

### EXPERIMENTAL PROCEDURES

To measure the absolute gain of the radio telescope, separate experiments were executed because of the need to be outside the Fraunhofer region, while still being able to receive the transmitted signal. The distance required by the far-field equation is 1664 m, which requires the remote transmitter; however, the dipole antenna could not receive a detectable signal at this distance. Therefore, it was determined that the relative gain would be found between the horn antenna and the radio telescope utilizing the remote transmitter at a distance greater than the 1664 m. Then the relative gain between the dipole and the horn antenna would be found using a HP frequency generator coupled to another dipole at a shorter distance; hence the three antenna method.

To determine the relative gain between the

horn antenna and the radio telescope a transmitter with dipole antenna (seen in Figure 3) was positioned on a local hillside that was approximately 2400 m away. The standard radio telescope's receiver was removed and replaced by a waveguide to coaxial transition used by the horn antenna. The waveguide to coaxial transition was attached to a spectrum analyzer with coaxial cable (shown in Figure 4). The telescope position was driven in azimuth and elevation until the maximum signal was received and the relative signal strength was noted. The waveguide to coaxial transition was then removed from the radio telescope and coupled to the horn antenna (Figure 5). The horn antenna's position was altered until the maximum signal strength was discovered and the relative signal strength was noted. This measurement process was completed in the following order (ABBABAAB) and is listed with the resultant values in Table 1. The measurements were taken in this order to allow for the removal of up to second order electronic drift. This measurement order was followed in order to remove both linear and qua-

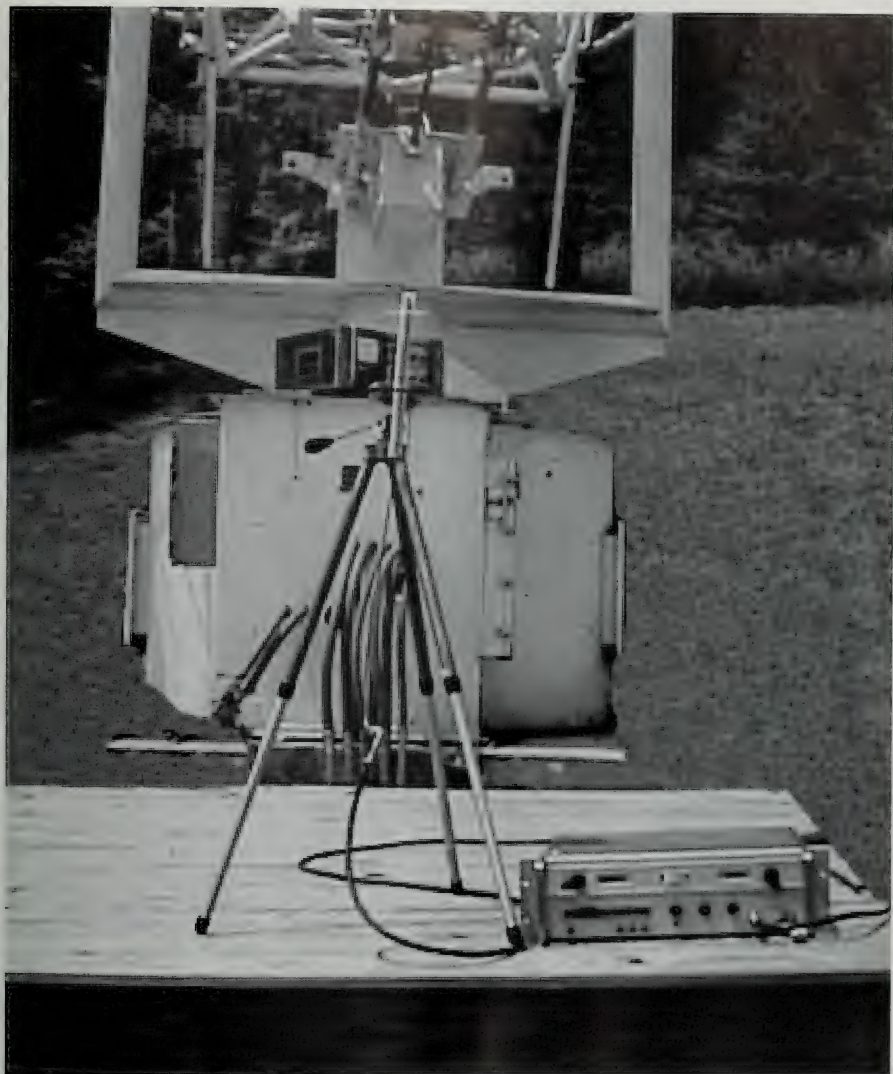


Figure 3. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. Experimental hardware used to determine the relative gain between the horn antenna and the radio telescope; shown is the commercial HP 8164A RF signal generator transmitter with the transmitting dipole antenna. The system was positioned on a local hillside ca. 2400 m away from the receiving antennas.



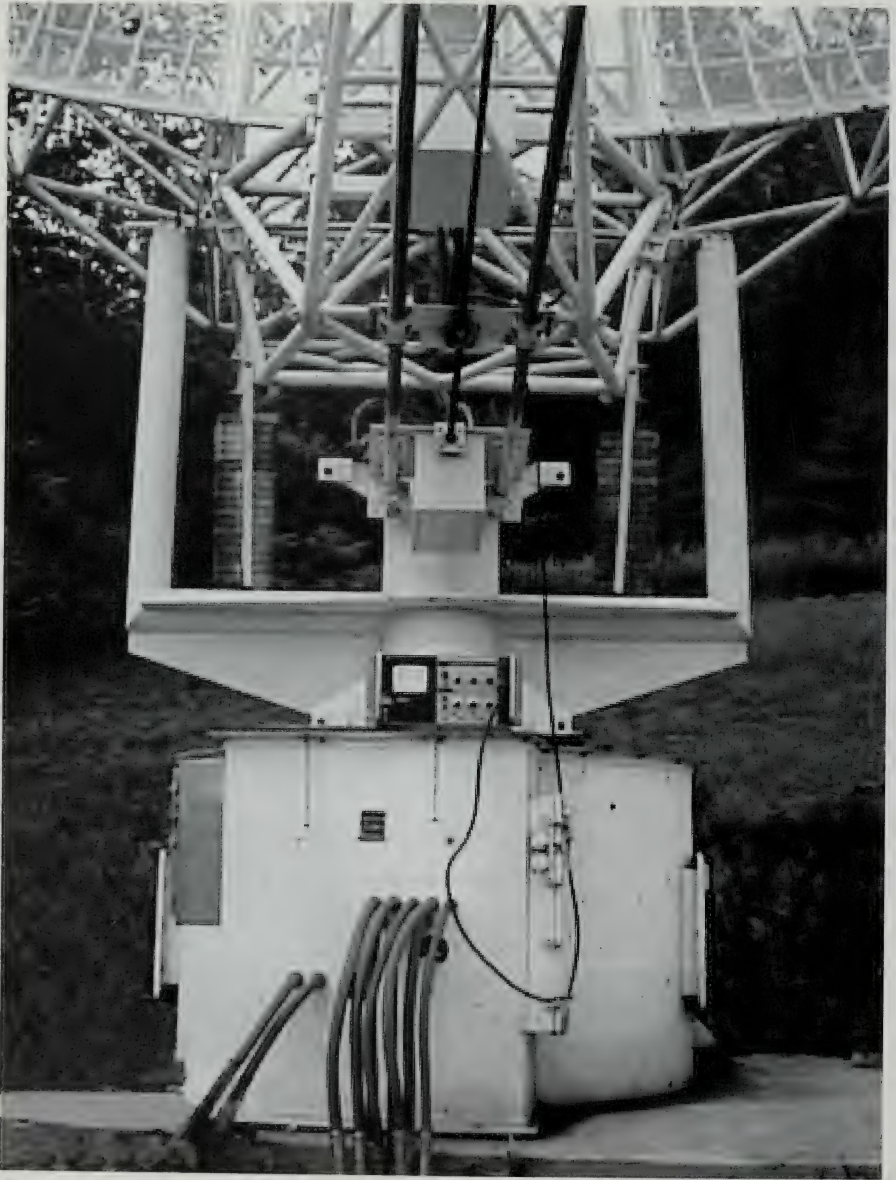


Figure 4. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. The telescope's standard receiver was removed and replaced by a waveguide to coaxial transition (also used by the horn antenna). The waveguide to coaxial transition was next attached to a IIP spectrum analyzer with coaxial cable. The system served as the receiving system for the MRT, horn antenna and dipole during the three-antenna method.



Figure 5. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. The horn antenna, waveguide to coaxial transition, and spectrum analyzer receiver system are shown. The horn antenna's position was altered until the maximum signal strength was discovered and the relative signal strength was measured.

dratic drifts in the electronics (i.e. frequency drifts in the transmitted signal). This experimental procedure yielded a result of  $24.9 \pm 1.2$  dBm relative gain between the horn antenna and the radio telescope.

Next the relative gain of the horn antenna

Table 1. Results of the empirical measurement of the horn antenna and the MRT, Morehead Radio Telescope, Morehead State University, Morehead, Kentucky.

Measurement	dBm Gain
Horn (A)	$\pm 46.0$
Telescope (B)	-19.9
Telescope (B)	-19.5
Horn (A)	-44.0
Telescope (B)	-20.0
Horn (A)	-44.0
Horn (A)	-45.0
Telescope (B)	-20.0

to the dipole antenna was calculated in the following manner. A transmitted test signal was produced with a HP frequency generator tuned to 1420 MHz coupled with a dipole antenna at a distance of 46 m from the horn antenna, well within the Fraunhofer pattern of the horn antenna, which is at a minimum distance of 8 m. The antenna range utilized met several desirable criteria. For example, the range was not a flat surface but had a several meter depression, ideal for avoiding reflection. The RF environment surrounding the antenna test range was also extremely low noise. A signal was transmitted and detected using the same experimental setup and methods as previously described. The position of the horn antenna was adjusted such that the strongest signal was detected and noted. The spectrum analyzer was attached to another dipole antenna

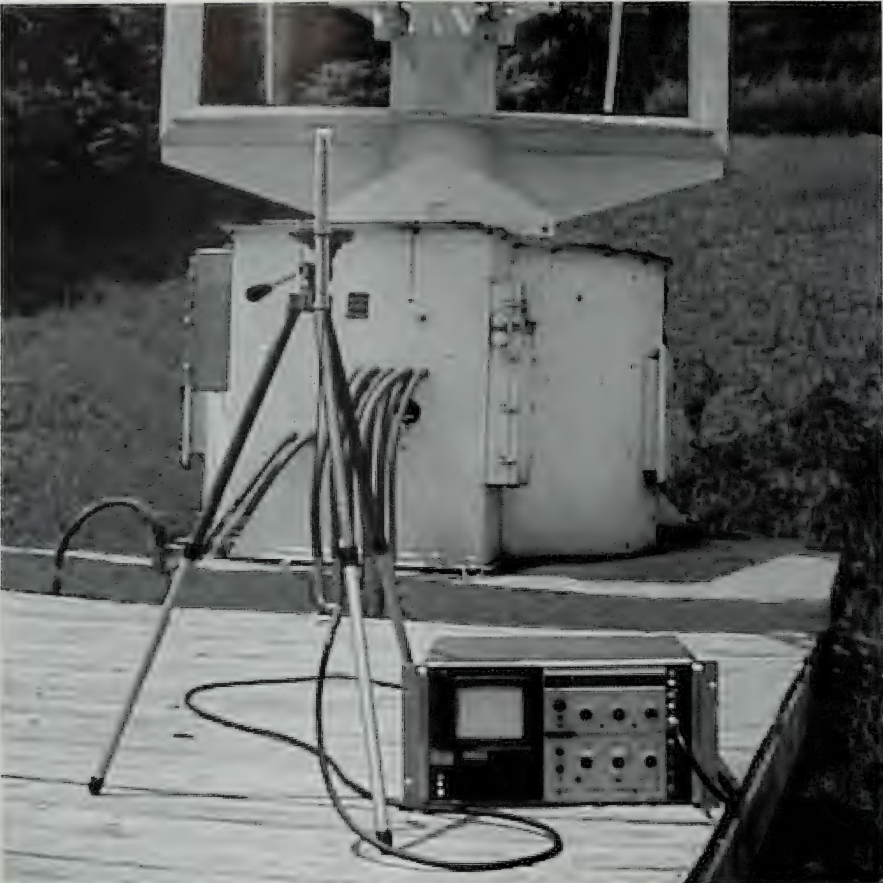


Figure 6. Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. The dipole antenna and spectrum analyzer receiver system are shown.

Table 2. Results of the empirical measurements of the horn and dipole antennae, Morehead Radio Telescope, Morehead State University, Morehead, Kentucky.

Measurement	dBm Gain
Horn (A)	-26.0
Dipole (B)	-41.5
Dipole (B)	-39.5
Horn (A)	-27.0
Dipole (B)	-41.0
Horn (A)	-26.0
Horn (A)	-26.5
Dipole (B)	-41.0

(Figure 6). The dipole was then positioned so that the maximum signal was received and noted. The data was taken in the same order as before, with the dipole taking the place of the radio telescope in the experimental order. This procedure yielded a result of  $14.4 \pm 1.3$  dBm. The measurement order and the recorded values are listed in Table 2.

Since the absolute gain of any dipole is 1.6 dB it follows that the gain of the MRT antenna is  $40.9 \pm 1.8$  dB. With this important result we can establish the performance characteristics of the MRT.



# PROJECT SIGNIFICANCE, PERFORMANCE CHARACTERISTICS, AND CONCLUSIONS

Although the gain of the antenna has been measured, there are still some very important characteristics of the antenna that need to be determined. The effective aperture,  $A_e$ , is perhaps one of the most important characteristics of an antenna. For a horn antenna,  $A_e$  is calculated using the dimensions of the mouth of the antenna. For other antennas, the calculation is much more cumbersome.  $A_e$  is related to the physical aperture of a parabolic dish by  $A_p = \pi D^2/4$ . The theoretical effective area of the MRT is 38 square meters. The effective area is related to the physical aperture by  $A_e = \eta A_p$ , where  $\eta$  is the aperture efficiency. The aperture efficiency can be calculated if the gain of the dish is known. The gain is simply the ratio of the signal received by the dish to that of an isotropic response. The effective area is calculated using equation 12

$$A_e = \frac{\lambda^2 G}{4\pi k_0} \quad (12)$$

Using our value for the gain and assuming a  $k_0$  (ohmic loss factor) of 1.5, we find a value of  $A_e = 29.1 \text{ m}^2$ . Given that the actual area is ca.  $38 \text{ m}^2$ , we estimate an aperture efficiency of 76%. This number is higher than expected because of inaccuracies (particularly time averaged) in the test equipment.

Using the effective aperture, the induced antenna temperature,  $T_A$ , can also be derived. For a source of known flux density ( $S$ ),  $T_A$  is simply the total power collected by the antenna divided by Boltzmann's constant,  $k$ . The expression for  $T_A$  is given below.

$$T_A = \frac{A_e S}{2k} \quad (13)$$

Using  $A_e = 29.1 \text{ m}^2$ , and assuming a flux of  $3.9 \text{ Jy}$  (the relevance of this number will soon be demonstrated) we find that  $T_A = 0.042 \text{ K}$  (Storey 1991).

The minimum detectable flux density,  $\Delta S_{\min}$  is another important antenna parameter. It is given by the equation

$$\Delta S_{\min} = \frac{2kK_s T_{\text{sys}}}{A_e \sqrt{\Delta \nu t n}} \quad (14)$$

where  $K_s$  is the sensitivity constant (ca. unity),

$T_{\text{sys}}$  is the total system temperature,  $\Delta \nu$  is the pre-detection bandwidth,  $t$  is the post-detection integration time, and  $n$  is the number of records averaged. Using previously determined values we found that  $\Delta S_{\min} = 3.9 \text{ Janskys}$  ( $10^{-26} \text{ Wm}^{-2} \text{ Hz}^{-1}$ ).

In the future we plan to measure the minimum detectable antenna temperature and compare it with results reported in the literature. We also plan a series of experiments to measure objects of known flux (flux calibrators) to determine the relationship between flux ( $S$ ) and induced antenna temperature ( $T_A$ ). Once this relationship is established, the induced antenna temperature during an observation can be converted to the objects actual integrated flux density. The measurements previously described are critical, therefore, in interpreting observations of cosmic phenomena with the MRT.

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## Notes on Nesting by the Smooth Softshell Turtle (*Apalone mutica*) in a River Impoundment in Western Kentucky

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### ABSTRACT

Smooth softshell turtles (*Apalone mutica*) in Kentucky Lake, an impoundment of the Tennessee River in western Kentucky and Tennessee, were observed to nest primarily on relatively flat beaches consisting of an upper layer of coarse gravel (several centimeters thick) that had been deposited by waves over sand, the turtle's preferred nesting substrate in free-flowing rivers. Nesting occurred throughout most of June and July. Four nests contained 15-20 eggs, and a dissected female contained 12 oviducal eggs and 13 enlarged follicles, for an overall average clutch size of 15.5. This average is intermediate to values for other populations from the northern half of the species' geographic range. There is only weak (marginally nonsignificant) evidence of a north-south cline in clutch size. Body size of the dissected female was comparable to those of the largest females captured in more extensive samples in two more northern populations. Egg diameter for two nests averaged 24.12 mm and 26.45 mm.

### INTRODUCTION

River impoundment is among the greatest anthropogenic influences affecting large-river ecosystems and their biotic communities (Dynesius and Nilsson 1994; Ward 1998). In Kentucky Lake, an impoundment of the lower Tennessee River in western Kentucky and Tennessee, one effect of impoundment is the loss of sandbars. For the smooth softshell turtle (*Apalone mutica*), a trionychid native to much of the Mississippi River drainage and several smaller drainages ranging from the western Florida panhandle to eastern Texas (Ernst et al. 1994), clean sandbars are preferred nesting habitat (Anderson 1958; Doody 1995; Fitch and Plummer 1975; Goldsmith 1944; Plummer et al. 1994; Webb 1962). Here I report on observations of nesting by *A. mutica* in Kentucky Lake.

### METHODS

I made fortuitous observations of nesting activity with a spotting scope in Nickell Cove, Kentucky Lake (Figure 1), during counts of basking turtles for a study of community ecology and longer-term observations of aggressive interactions of basking turtles (Lindeman 1999, 2000). I excavated eggs from nests for

enumeration, and measured egg diameters in two clutches with vernier calipers. Additional reproductive data were taken from a female *A. mutica* that drowned after a trap was pulled into deep water by a beaver.

### RESULTS

On 8 Jun 1993 I observed two *A. mutica* nesting, one from 1009 to 1055, the other from 1058 to 1128. Excavations revealed 15 and 16 eggs, respectively, in nests at the crest of the beach (hereafter Beach 1) less than 2 m from the water's edge. Beach 1 was ca. 60 m long, less than 1 m above the water's surface, and from 1-3 m wide. Eggs were surrounded by sand, but to reach the sand, females had to dig through 10-15 cm of flat, coarse gravel up to 5 cm in diameter that had accumulated on the beach due to wave action. With the females facing away from the water, digging was done with the hind feet such that gravel was thrown toward the water. Several other *A. mutica* were basking at the water's edge; on one occasion the flying gravel resulted in a male *A. mutica* abandoning its basking position and seeking shelter in the water.

On 15 Jun 1994 at 1000 I observed a female nesting on the opposite side of the cove, about 1 m from water on the leeward tip of a flat, semicircular beach (Beach 2) that was 40 m long, up to 20 m wide, and ca. 1.5 m above the water's surface. Most of this beach was covered with flat, coarse gravel deposited by

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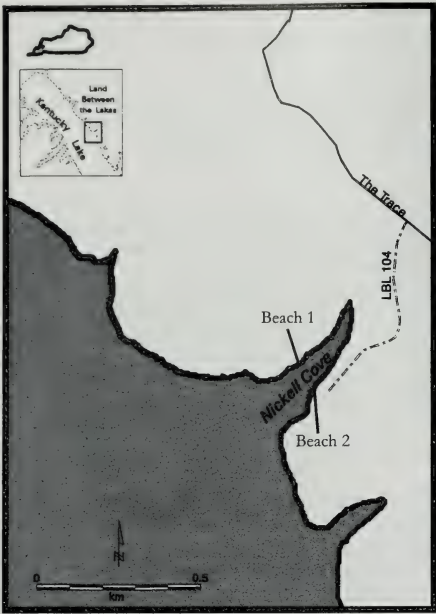


Figure 1. Location of Nickell Cove in Kentucky Lake, Kentucky.

wave action, but on the date of nesting the leeward tip had an area of about 6 m<sup>2</sup> that was primarily sand, and the nest was constructed in sand. Twenty eggs were laid, and averaged 24.12 mm in diameter (range 23.3–25.0 mm). On 6 Jul 1994 I observed a female nesting on Beach 1 between 0945 and 1015. Excavation revealed 17 eggs in sand below several cm of gravel. Two eggs were damaged by gravel during excavation; the remaining 15 eggs av-

eraged 26.45 mm in diameter (range 25.4–27.9 mm). On 20 Jul 1994 I observed a female digging in gravel on Beach 1 at 1000, but she fled into the water upon my approach. On 12 Jul 1996 I observed a female digging in gravel on Beach 1 at 1145. Subsequent attempts to find a nest were unsuccessful, and oviposition may not have occurred. On 13 Jul 1996 a female (straight-line carapace length [CL] 299 mm, plastron length [PL] 209 mm) was drowned in a trap set adjacent to Beach 1. Dissection revealed 12 shelled oviducal eggs and 13 large-diameter (16–20 mm) follicles that presumably represented another clutch of eggs that would have been laid that year.

DISCUSSION

Reproductive data for *A. mutica* from Kentucky Lake are comparable to those in reports from elsewhere. Mean clutch size (15.5 eggs) is intermediate to other values for the species between 35°N and 43°N latitude, while clutch size declines sharply further south (Table 1). However, whereas clutch size tends to increase steadily with latitude in a variety of North American emydrid turtles (Iverson and Smith 1993; Litzgus and Brooks 1998; Seigel 1980; Tucker et al. 1998) and in wide-ranging kinosternid (Tinkle 1961) and chelydrid turtles (Iverson et al. 1997), and has been suggested to do so for *A. mutica* (Fitch 1985), the correlation of clutch size with latitude was positive but not significant across eight populations (Figure 2;  $r = 0.67$ ,  $P = 0.072$ ). In other North American freshwater turtle species, the larger clutch sizes of northern populations are usually associated with larger

Table 1. Reports of clutch size in *Apalone mutica* from across its geographic range.

Latitude	State(s)	Average clutch size (range, N)	Source
42°40'N	South Dakota	13.1 (9–16, 18)	Timken 1968
42°30'N	Iowa	18.7 (10–31, 16)	Goldsmith 1944
41°30'N	Illinois	18.4 (4–33, 8)	Muller 1921
41°20'N	Iowa	13.4 (7–19, 20)	Janzen 1993
39°00'N	Kansas	10.4 (3–26, 102)	Plummer 1977
36°55'N	Kentucky	15.5 (12–20, 6)	Present study
35°10'N	Arkansas	14.5 (8–23, 34)	M. Plummer, pers. comm.
30°30'N	Louisiana	6.7 (2–12, 55)	Doodly 1995
S. of 36°30'N	Various	7.3 (3–15, 12)	Webb 1962
S. of 36°30'N	Oklahoma & Texas	9.0 (3–25, 9)	Fitch 1985

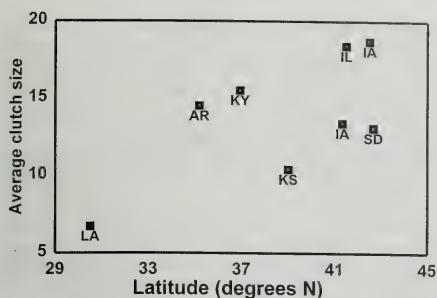


Figure 2. Relationship of clutch size to latitude for eight populations of *Apalone mutica*. State codes are given below each data point (see Table 1).

body sizes (Iverson and Smith 1993; Iverson et al. 1997; Litzgus and Brooks 1998; Tinkle 1961). It is thus noteworthy that my single Kentucky specimen (299 mm CL, 209 mm PL) exceeded by 15% the largest of 30 mature females captured by Timken (1968; 261 mm CL) 5°45' further north in South Dakota, and by 12% the largest of 173 mature females captured by Plummer (1977; 186 mm PL) 2°05' further north in Kansas. In addition, one of two adult female paratypes of *A. m. calvata*, from Escambia County, Florida, was 263 mm CL and 180 mm PL (Webb 1959), nearly identical to the largest individuals from the South Dakota and Kansas samples. It thus appears that *A. mutica* may have no latitudinal trend in body size and at best a weak latitudinal trend in clutch size. Additional data from southern populations will be necessary to fully evaluate these relationships.

The nesting season for *A. mutica* in Kentucky Lake appears to span most of June and July. Doody (1995) reviewed reports of nesting activity across the range of the species and found nesting activity to occur as early as 17 May and as late as 23 July. Mean egg diameter in Kentucky (25.12 mm) is slightly greater than values in the literature (22.6, Cahn 1937; 22.9 mm, Fitch and Plummer 1975; ca. 23 mm, Muller 1921; 23 mm, Timken 1968). Previous reports do not include the range of egg size and how it varies among clutches and thus the larger value for Kentucky may simply be an artifact of having data from only two clutches.

The unique aspect of my observations of re-

production in *A. mutica* in Kentucky Lake concerns the nesting substrate. The loss of sandy areas for nesting has been raised as a concern for trionychid turtles inhabiting reservoirs (Taskavak and Atatiir 1998). In Kentucky Lake, females must dig through coarse gravel to reach a sandy substrate. This change in nesting substrate may have important implications for embryonic development, nest predation, and success of emerging hatchlings. A study of the survivorship of eggs and emerging hatchlings of *A. mutica* nests on gravel beaches of reservoirs would be of great interest.

#### ACKNOWLEDGMENTS

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## Turtle Fauna of the Upper Tradewater River Near Dawson Springs, Kentucky

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### ABSTRACT

Published dot-distribution maps for aquatic turtles native to western Kentucky indicate a dearth of data from the region of the Tradewater River. I recorded eight species by using a spotting scope and trapping at six localities on the upper Tradewater River near Dawson Springs, Kentucky, and present data on their relative abundance in basking counts and captures.

### INTRODUCTION

Management and conservation of wildlife require first and foremost accurate information regarding distribution and historical abundance of species. Gibbons et al. (1997) discussed the importance of data on the geographic distribution of reptiles and amphibians and the value of data collected over a historical time frame. These animals can generally be handled and closely examined to ensure species identification. In addition, many aquatic turtles are easily observed because of their basking habits (e.g., Lindeman 1996, 1997, 1999).

Distributions of reptiles and amphibians are traditionally represented with dot-distribution maps plotting the collecting localities of museum specimens or sight records. No such maps have been published specifically for the turtles of Kentucky, although Iverson's (1992) range maps include Kentucky localities for the 12 freshwater species ranging into the state; only the painted turtle *Chrysemys picta* is shown as having been recorded in the Tradewater River area. Here I report on observations of the Tradewater River turtle fauna made in 1994-1999.

### METHODS

The Tradewater River drains portions of Christian, Hopkins, Caldwell, Crittenden, Webster, and Union counties in western Kentucky northward toward its confluence with the Ohio River. Six sites on the upper Tradewater River near Dawson Springs, Kentucky, were visited sporadically during warm weather (between 18 Mar and 15 Oct) in 1994-1995 and 1998-1999 (Figure 1). The sites were as-

sociated with roadway access: (A) Hopkins Park Road, (B) McKnight Road, (C) Old Hospital Road, (D) Old Mill Dam Road, (E) State Highway 62, and (F) State Highway 1220. The Tradewater River in this area is a sluggish, muddy stream with forested banks, depths seldom exceeding 3 m, abundant submerged and emergent deadwood, few aquatic macrophytes, and numerous rocky outcroppings along the banks.

I observed turtles by using a spotting scope with 22-60 $\times$  zoom magnification and trapped with fykenets (Vogt 1980) and floating basking traps (MacCulloch and Gordon 1978). Captured turtles were measured, marked with shell notches, and released at the site of capture. Following standard practice in turtle studies, I report midline plastron lengths (PL) for emydid turtles and midline carapace lengths (CL) for all other species.

### RESULTS AND DISCUSSION

Counts of basking turtles are summarized in Table 1; trapping results, in Table 2. Comments on the eight species I recorded in the Tradewater River and on four other aquatic turtles native to western Kentucky that I did not record follow.

#### Emydidae

The slider turtle, *Trachemys scripta*, the most abundant species sampled in spotting-scope counts and trapping, was a major component of the turtle fauna at all sites. This species is typically abundant in other localities in western Kentucky (Lindeman 1997, 2000). Of 21 captured *T. scripta* that were measured, 8 were males averaging 162.6 mm PL (range

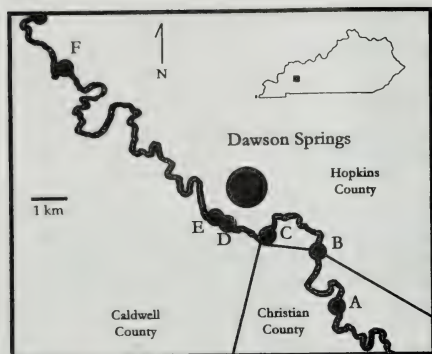


Figure 1. Map of the Tradewater River near Dawson Springs, Kentucky, showing the six survey sites (letters; see text for identification).

135–183 mm), 11 were females averaging 187.5 mm PL (range 125–214 mm), and 2 were unsexed juveniles (93 and 103 mm PL).

I observed the river cooter, *Pseudemys concinna*, frequently at sites C (40% of all turtles) and E (31% of all turtles). Compared to results obtained by Lindeman (1997) for nearby Kentucky Lake reservoir, these percentages are high. I captured a single female (176 mm PL) at site E.

The common map turtle, *Graptemys geographica*, appeared to be the turtle species with the most localized distribution within the upper Tradewater River. It was the most abundant turtle seen and trapped at site A but was only rarely recorded at other sites. I measured two of three captured turtles, a male (89 mm PL) and an unsexed juvenile (76 mm PL). I

saw the Ouachita map turtle, *G. ouachitensis*, twice at site E, but these observations may have been of the same turtle, as both involved males basking on the same log 2 days apart. I did not record the false map turtle, *G. pseudogeographica*. In Kentucky Lake reservoir and the lower Tennessee River, *G. ouachitensis* and *G. pseudogeographica* are common and *G. geographica* is very rare (Lindeman 1999). Fuselier and Edds (1994) described habitat partitioning among these three species in southeastern Kansas where *G. geographica* was more likely than the other two species to enter small streams. A similar partitioning of habitat in western Kentucky seems likely.

I did not record the painted turtle, *Chrysemys picta*, although Iverson (1992) showed the species as present in the area. This species, the most geographically widespread in North America, is abundant throughout most of its range (Ernst et al. 1994). Although it can be common in riverine habitats in the northern portion of its range (e.g., MacCulloch and Secoy 1983), in the southern portion of its range it is probably more restricted to lentic waters (Mount 1975). I have not found it to be abundant at any location in western Kentucky (P. Lindeman, unpubl. observ.).

#### Kinosternidae

The common musk turtle, *Sternotherus odoratus*, was observed basking at sites A, D, and E and trapped at sites B and E. It is probably a more abundant species than these results indicate, as it basks only rarely (Lindeman 1996). Four captured males averaged 90.5 mm CL (range 78–106 mm CL), and a

Table 1. Summary of basking turtles observed at four sites (Figure 1; see text for exact locations) on the upper Tradewater River, Kentucky, 1994–1999. Numbers of observations are given with percentages in parentheses.

Species	Site				Total
	A	C	D	E	
<i>Trachemys scripta</i>	33 (20)	50 (53)	3 (50)	45 (58)	131 (38)
<i>Pseudemys concinna</i>	1 (<1)	38 (40)	1 (17)	24 (31)	64 (19)
<i>Graptemys geographica</i>	113 (68)	1 (1)	0	0	114 (33)
<i>G. ouachitensis</i>	0	0	0	2 (3)	2 (1)
<i>Sternotherus odoratus</i>	1 (<1)	0	1 (17)	1 (1)	3 (1)
<i>Kinosternon subrubrum</i>	1 (<1)	0	0	0	1 (<1)
<i>Chelydra serpentina</i>	1 (<1)	0	0	0	1 (<1)
<i>Apalone spinifer</i>	10 (6)	1 (1)	0	4 (5)	15 (4)
Unidentified turtles	6 (4)	4 (4)	1 (17)	2 (3)	13 (4)
Total turtles	166	94	6	78	344
Total survey days	39	31	5	18	

Table 2. Summary of trapping effort, types of traps used, and turtles trapped at four sites (Figure 1; see text for exact locations) on the upper Tradewater River, Kentucky, 1994–1999. Numbers trapped are given with percentages in parentheses.

Species	Site				Total
	A	B	E	F	
<i>Trachemys scripta</i>	1 (25)	1 (50)	10 (50)	11 (85)	23 (59)
<i>Pseudemys concinna</i>	0	0	1 (5)	0	1 (3)
<i>Graptemys geographica</i>	2 (50)	0	0	1 (8)	3 (8)
<i>Sternotherus odoratus</i>	0	1 (50)	4 (20)	0	5 (13)
<i>Chelydra serpentina</i>	0	0	3 (15)	1 (8)	4 (10)
<i>Apalone spinifera</i>	1 (25)	0	2 (10)	0	3 (8)
Total turtles	4	2	20	13	39
Total trap nights:					
Fykenets	1	8	11	10	30
Basking traps	4	0	12	2	18

captured female was 85 mm CL. A single common mud turtle, *Kinosternon subrubrum*, was observed basking at Site A. Collins (1995) recorded a carcass of *K. subrubrum* from site E. This species also basks only rarely (Lindeman 1996) and thus may be more abundant than is indicated.

### Chelydridae

I recorded the common snapping turtle, *Chelydra serpentina*, at sites A, E, and F. The rarity with which this species basks (Ernst et al. 1994) makes it unlikely to be recorded in basking surveys. I measured only one of four turtles captured, a male (326 mm CL). I did not record the alligator snapping turtle *Macrochelys temminckii*; Iverson's (1992) range map indicates that the Tradewater River is probably just outside the turtle's range.

### Trionychidae

The spiny softshell turtle, *Apalone spinifera*, was seen basking at sites A, C, and E and captured at sites A and E, but was nowhere abundant. Captured individuals included a male (151 mm CL) and two females (292 and 405 mm CL), the larger of which was within 27 mm of the record size recorded for the eastern subspecies, *A. s. spinifera* (432 mm CL; Conant and Collins 1998). The smooth softshell turtle, *A. mutica*, was not recorded and probably does not occur in the upper Tradewater River as it is known to be more restricted to larger rivers than is *A. spinifera* (Williams and Christiansen 1981).

My surveys, together with the dot distribu-

tion map of Iverson (1992) for *Chrysemys picta*, indicate the presence of nine of western Kentucky's 12 aquatic turtle species in the Tradewater River drainage; the remaining three Kentucky species are unlikely to occur in the area. The drainage is thus an important area for turtle diversity within the state. The data presented here may be valuable for future assessment of the impacts of agricultural runoff, erosion, surface mining, and other forms of water-quality degradation on turtle assemblages.

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# Macroinvertebrate, Fish, and Physicochemical Differences Between an Acid Mine Drainage Impacted Stream and a Kentucky Wild and Scenic River

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## ABSTRACT

Acid mine drainage (AMD) causes a myriad of impacts to macroinvertebrates and fishes and causes changes in the chemical properties of water. During 1991 and 1992 macroinvertebrate, fish, and physicochemical data were collected and compared from two Kentucky watercourses: Bear Creek, an AMD impacted stream, and Rock Creek, a Kentucky Wild and Scenic River. Macroinvertebrates, fishes, and physicochemical data were also collected from Bear Creek in 1994 to determine if reclamation efforts had been successful. Three replicate 0.09 m<sup>2</sup> Surber samples (900  $\mu$ m mesh) supplemented with qualitative sampling in available habitats were used for macroinvertebrate sampling. Fishes were collected with a 3.4  $\times$  1.2 m seine (0.3 cm mesh) in all available habitats. Water samples for physicochemical analyses were collected with either grab or depth-integrated techniques. Significant differences (Mann-Whitney U,  $P \leq 0.05$ ) were found between the macroinvertebrate taxa richness, total number of individuals, EPT, %EPT, %Chironomidae and mHBI values; the fish IBI values; and 13 of the 25 physicochemical variables examined from the streams. Reclamation occurred at three locations in the Tennessee portion of the Bear Creek watershed, but no improvements in water quality in the Kentucky portion were observed.

## INTRODUCTION

The extraction of coal has occurred in the United States for well over 200 years (Parsons 1968) and negatively affects aquatic systems primarily through sedimentation (Waters 1995) or acid mine drainage (AMD) (Earle and Callaghan 1998; Starnes and Gasper 1996). AMD causes a myriad of impacts to macroinvertebrates and fish, including physical (e.g., covering of gills) and physiological (e.g., disruption of ion transport mechanisms) effects (Earle and Callaghan 1998), changes in community composition and structure (Branson et al. 1984; Canton and Ward 1981; Parsons 1968), and destruction of habitat (e.g., deposition of Fe(OH)<sub>3</sub>) (Moon and Lucostic 1979). AMD also causes changes in the chemical properties of water, such as increases in several heavy metals (e.g., Fe and Al) and a decrease in the ability of water to ameliorate adverse pH (Becker et al. 1986; Parsons 1968; Rikard et al. 1986). Complete discussions on

the formation and impacts of AMD can be found in Earle and Callaghan (1998), Hynes (1960), and Parsons (1968).

The present study is part of an AMD non-point source interstate demonstration project originally coordinated between the Commonwealth of Kentucky and the state of Tennessee. One objective of the project was to collect and compare macroinvertebrate, fish, and physicochemical data from AMD-impacted Bear Creek and a relatively undisturbed section of Rock Creek. A second objective was to assess the success of reclamation activities in the Bear Creek watershed (KDOW 1999; Stucki 1995).

## STUDY AREA

Both study streams are tributaries of the South Fork Cumberland River (SFCR). A large portion of the SFCR watershed is classified and operated by the National Park Service as the Big South Fork National River and Recreation Area (BISO). The waterways feed-

ing the BISO vary in their degree of quality, and are impacted by agriculture, coal mining, oil and gas exploration, sewage discharge and silviculture (Rikard et al. 1986). Bear Creek is the most severely AMD-impacted stream in the upper Cumberland River basin, as evidenced by high conductivity levels, low pH, increased sedimentation, and the presence of other pollutants (Rikard et al. 1986; Stucki 1995; USDA 1991).

The two forks of Bear Creek originate near Oneida, Tennessee, and converge to form the Bear Creek mainstem ca. 1.6 km south of the Kentucky/Tennessee state line. Bear Creek flows in a northwesterly direction until its confluence with SFCR (at SFCR km 81.4) (Alexander and Robison 1997; Stucki 1995). The Bear Creek watershed encompasses ca. 60.5 km<sup>2</sup> (Bower and Jackson 1981) and includes portions of Scott County, Tennessee, and McCreary County, Kentucky (Alexander and Robison 1997; Stucki 1995).

Extraction of minerals within the headwaters of Bear Creek began in the late 1800s and continued sporadically but was non-existent during passage of the Surface Mining Reclamation and Control Act (P.L. 95-87) in 1977 (Alexander and Robison 1997). About 33 sites (USDA 1991) encompassing 310 ha of abandoned mine lands and reclaimed lands (Alexander and Robison 1997) exist in the watershed. The majority of land within the Bear Creek watershed is forested (81% or 2,833 ha). The remaining land uses are mining (7%), grassland (8%), urban (3%), and cropland (1%) (USDA 1991).

Rock Creek originates in Fentress County, Tennessee, flows northeastward through Pickett and Scott counties, Tennessee, and into the SFCR at Yamacraw, McCreary County, Kentucky (SFCR km 65.8) (Harker et al. 1980; Layzer and Anderson 1992). The Rock Creek watershed encompasses ca. 162.2 km<sup>2</sup> (Bower and Jackson 1981), and in Kentucky is almost entirely located within Daniel Boone National Forest. The watershed is ca. 95% forested (Harker et al. 1980), with only a small amount of land devoted to agriculture.

From the Kentucky/Tennessee state line (km 35.2) to the confluence with White Oak Creek (km 6.3), Rock Creek has been designated a Kentucky Wild River since 1972 (Miller, Wihry, and Lee, Inc. 1980). This segment

of Rock Creek is also under consideration as a National Wild and Scenic River (M. Jones, Kentucky Division of Water, pers. comm., 26 Sep 2000). Below this confluence, however, Rock Creek is severely impacted by AMD from White Oak Creek (Layzer and Anderson 1992; Rikard et al. 1986).

Both study streams are located within the Cumberland Plateau Section of the Central Appalachian Ecoregion (Burr and Warren 1986; Omernik 1987), in the southern portion of the Appalachian Plateaus Physiographic Province. The topography of this section is extremely rugged, with elevations ranging between 366 and 549 meters; streams in this section have moderate to high gradients, with poorly developed floodplains (Burr and Warren 1986). Four major river basins (Big Sandy, Cumberland, Kentucky, and Licking River) drain the Central Appalachian Ecoregion. It is heavily forested and underlain by Pennsylvanian age strata (KDOW 1997) that include sandstone, siltstones, and coal. One monitoring station was established near the mouth of Bear Creek (36°37'32.3"N, 84°31'58.2"W) and another in Rock Creek at the Kentucky/Tennessee state line (36°36'8.5"N, 84°44'24.0"W) (Figure 1).

## MATERIALS AND METHODS

### Macroinvertebrates

A 0.09 m<sup>2</sup> Surber sampler (900 µm mesh) was used for quantitative macroinvertebrate sampling. Three replicate samples were taken from comparable riffle substrate at each station. For qualitative sampling, three standard efforts (jabs, kicks, sweeps, etc.) were performed in available habitat with D-frame dip nets (800 × 900 µm mesh). Macroinvertebrates were collected during winter (Feb), spring (Apr), summer (Jul), and fall (Oct) at both sites from 1991 to 1992. The Bear Creek site was also sampled for macroinvertebrates during fall 1994 to determine if reclamation efforts initiated in 1992 in the Tennessee portion of the watershed (Stucki 1995) had been successful. All macroinvertebrate samples are deposited in the Branley A. Branson Museum of Zoology, Eastern Kentucky University.

Macroinvertebrates were identified to the lowest possible taxonomic level, usually genus or species, and were evaluated with richness,



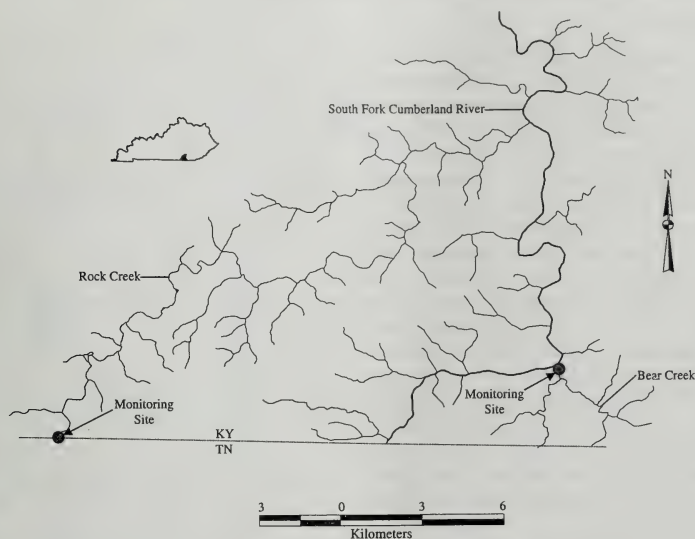


Figure 1. Bear Creek and Rock Creek monitoring locations. Inset shows the location of McCreary County, Kentucky.

composition, and tolerance measures (metrics). Richness measures included taxa richness (s), total number of individuals (TNI), and number of mayfly (Ephemeroptera), stonefly (Plecoptera), and caddisfly (Trichoptera) taxa (EPT). Composition measures included the percentage of each community represented by EPT (%EPT) and midges (Diptera: Chironomidae) (%CHIR) (Barbour et al. 1999). A modified Hilsenhoff Biotic Index (mHBI) (i.e., the North Carolina Biotic Index, Lenat 1993) was used to establish the overall pollution tolerance of each macroinvertebrate community.

## Fishes

Fishes were collected at each site for 0.5 to 1.0 hr with a minnow seine ( $3.4 \times 1.2$  m, 0.3 cm mesh) in all available habitats at each site during spring and fall 1991 and 1992. Fishes were collected from Bear Creek during fall 1994 to determine if reclamation efforts had been successful and from Rock Creek for comparison. Fishes were identified to species. All fish samples are deposited in the Branley A. Branson Museum of Zoology, Eastern Kentucky University. The Index of Biotic Integrity (IBI) (Karr 1981) was used to determine the

overall health of the fish community using tentative scoring criteria established for the Central Appalachian Ecoregion (M. Compton, Kentucky Division of Water, pers. comm., 30 Mar 2001).

## Physicochemical Variables

High flow and normal/low flow water samples for physicochemical analyses were collected during 1991 and 1992 at both stations ( $n_{\text{Bear Creek}} = 15$ ;  $n_{\text{Rock Creek}} = 8$ ) with either grab or depth-integrated samples, or with an automated sampler, depending on rainfall amounts and flow conditions. Water samples ( $n = 2$ ) were also collected from Bear Creek during fall 1994 to assess reclamation efforts. Temperature, pH, dissolved oxygen (D.O.), specific conductivity, and turbidity were measured in situ with portable meters. Water samples were placed into containers, preserved with  $\text{H}_2\text{SO}_4$  (if necessary), and placed on ice until delivery to the laboratory. Twenty variables—Al, Ba, Ca, Co, Cr, Cu, Fe, Hg, Mg, Mn, Na, Ni, K, Zn,  $\text{Cl}^-$ ,  $\text{SO}_4$ , alkalinity, hardness, total suspended solids (TSS), and total volatile solids (TVS)—were analyzed for each sample following standard methods (e.g., APHA et al. 1998).

Data Comparison

The non-parametric Mann-Whitney U test (StatMost, ver. 2.01, DataMost Corporation) was used to compare the macroinvertebrate, fish, and physicochemical data from the two streams and to compare pre- and post-reclamation data from Bear Creek. Macroinvertebrate communities were also compared using the Community Similarity Index ( $PS_c$ ) (Krebs 1989). For the pre- and post-reclamation comparisons of macroinvertebrate and fish data, the fall 1994 collections were compared to the fall 1991 and 1992 collections to avoid seasonal influences. The post-reclamation physicochemical data was compared to all of the pre-reclamation data. Non-detect physicochemical values (i.e., those less than the established detection limit for each variable) were assumed to be equal to zero, which may have introduced bias to the statistical analyses (Travis and Land 1990).

RESULTS

Faunal and Physicochemical Comparisons

**Macroinvertebrates.** A total of 517 individuals representing 81 taxa were collected from Bear Creek during 1991 and 1992 (Appendix A). Bear Creek taxa richness and TNI fluctuated throughout the study, with the highest values during summer 1991 (39 and 182, respectively). The lowest taxa richness value (10) occurred during winter 1991 and summer 1992, and the lowest TNI value (16) occurred during winter 1991. The average EPT value was 4.9, with values ranging from 1 (winter 1991) to 12 (fall 1991) (Table 1). With the exception of spring (33.3%) and fall (59.5%) 1992, the Bear Creek %EPT values were below 25%. The Bear Creek %CHIR values fluctuated from 9.5% (spring 1991) to 72.4% (summer 1992) and averaged 31.8%. The average Bear Creek mHBI was 5.75, with values ranging from 4.04 (summer 1991) to 7.01 (spring 1991).

A total of 4658 individuals representing 143 taxa were collected from Rock Creek during 1991 and 1992 (Appendix A). Rock Creek taxa richness and TNI also fluctuated throughout the study; the highest values (77 and 1084, respectively) were obtained during summer 1991. The lowest taxa richness value (37) occurred during winter 1991 and 1992; the low-

Table 1. Macroinvertebrate and fish metric values for Bear Creek (BC) and Rock Creek (RC), McCreary County, Kentucky, during winter [win], spring [spr], summer [sum] and fall, 1991-1992.

Metric	Win 91		Spr 91		Sum 91		Fall 91		Win 92		Spr 92		Sum 92		Fall 92		Fall 94	
	BC	RC	BC	RC	BC	RC	BC	RC	BC	RC	BC	RC	BC	RC	BC	RC	BC	RC
s	10	37	14	46	39	77	38	70	22	37	17	65	10	46	17	49	16	—
TNI	16	691	21	333	182	1084	106	1057	41	258	42	446	29	310	42	465	40	—
EPT	1	21	2	27	5	30	12	33	4	20	5	27	3	18	8	24	4	—
%EPT	6.2	13.9	14.2	82.6	7.1	47.4	16.9	68.8	14.6	49	33.3	65	10.3	45.8	59.5	63.6	20	—
%CHIR	43.8	18.2	9.5	6.6	41.8	39.6	31.1	14.8	39	6.2	14.3	13.2	72.4	11.3	2.4	8	7.5	—
mHBI	5.46	4.14	7.01	2.01	4.04	4.34	6.47	3.61	6.61	3.64	5.96	3.00	5.87	3.69	4.56	3.81	7.39	—
$PS_c$	—	—	11.0	—	18.7	—	9.2	—	9.4	—	7.8	—	5.7	—	6.7	—	—	—
IBI	—	—	0	26	—	—	8	26	—	—	0	28	—	—	0	38	0	34

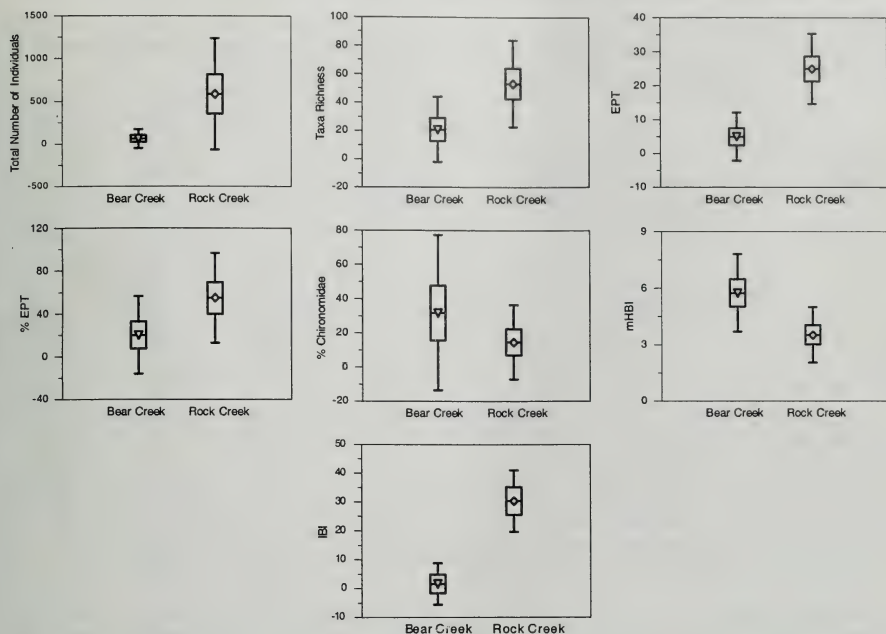


Figure 2. Comparison of macroinvertebrate and fish metrics from Bear Creek and Rock Creek, McCreary County, Kentucky, 1991–1992. Bars on the plots correspond to, in increasing order, mean  $\pm$  2  $\times$  standard deviation, mean  $\pm$  2  $\times$  standard error, mean, mean  $\pm$  2  $\times$  standard error and mean  $\pm$  2  $\times$  standard deviation.

est TNI during winter 1992 (288). Rock Creek EPT values ranged from 18 (summer 1992) to 33 (fall 1991), with an average value of 25 (Table 1). The Rock Creek %EPT values, with the exception of winter 1991 (13.9%), were relatively high (%EPT  $\geq$  45). The %CHIR values were relatively low, ranging from 6.2% (winter 1992) to 39.6% (summer 1991). The average Rock Creek mHBI was 3.53, with values ranging from 2.01 (spring 1991) to 4.34 (summer 1991).

The macroinvertebrate  $PS_c$  values ranged from 1.2 (winter 1991) to 18.7 (summer 1991), with an average value of 8.7 (Table 1) that indicated a large degree of dissimilarity between the two streams. Rock Creek had significantly higher ( $P \leq 0.05$ ) taxa richness, TNI, EPT, and %EPT values, and significantly lower ( $P \leq 0.05$ ) %CHIR and mHBI values (Figure 2).

**Fishes.** Attempts were made to collect fish at the Bear Creek station during spring and fall 1991 and 1992. Only one fish was collected during these four attempts (Table 2). With the

exception of the fall 1991 collection (IBI = 8), the resulting IBI values for all of the collections were 0 (Table 1). All of the Bear Creek fish collections had a water quality rating of "Very Poor."

The four collections of fish from Rock Creek yielded 907 individuals, representing 6 families and 19 species (Table 2). The IBI values for Rock Creek ranged from a low of 26 ("Fair" water quality) during spring and fall 1991 to a high of 38 ("Excellent" water quality) during fall 1992. The average IBI value for the five collections was 30.4 (Table 1), indicating an overall water quality rating of "Good." Rock Creek IBI scores were significantly ( $P \leq 0.05$ ) higher than Bear Creek IBI scores (Figure 2).

**Physicochemical variables.** Several physicochemical variables from Bear Creek had substantial variation (standard deviation  $\geq$  mean) (Table 3). For example, alkalinity ranged from 0.0 (not detected) to 11.4 mg/liter ( $\bar{x} = 1.66 \pm 2.78$  mg/liter), and Cr ranged



Table 2. Fishes collected from Bear Creek (BC) and Rock Creek (RC), McCreary County, Kentucky, during 1991, 1992 and 1994. Values are presented as BC/RC, during spring [spr] and fall. A blank space indicates the organism was not collected.

Taxa		1991		1992		1994
		Spr	Fall	Spr	Fall	Fall
Petromyzontidae	Lamprey larvae	-/1			-/1	
Cyprinidae	<i>Camptostoma anomalum</i>		-/3	-/3	-/6	-/3
	<i>Cyprinella galacturus</i>		1/-			
	<i>Cyprinella spiloptera</i>			-/1		
	<i>Lythrurus fasciolaris</i>	-/21	-/27		-/2	-/48
	<i>Notropis amblops</i>	-/6		-/45	-/30	-/7
	<i>Notropis arionunus</i>			-/115		
	<i>Notropis galacturus</i>	-/1			-/7	
	<i>Notropis telescopus</i>	-/34	-/78	-/81	-/91	-/84
	<i>Notropis volucellus</i>		-/70	-/214	-/20	-/177
	<i>Semotilus atromaculatus</i>			-/5		-/5
Catostomidae	<i>Hypentelium nigricans</i>		-/1		-/1	
	<i>Moxostoma duquesnei</i>				-/1	
Ictaluridae	<i>Noturus flavus</i>				-/1	-/1
Centrarchidae	<i>Micropterus salmoides</i>	-/1				-/1
Percidae	<i>Etheostoma blennioides</i>		-/3	-/3	-/1	-/2
	<i>Etheostoma caeruleum</i>	-/1	-/5	-/5	-/12	-/22
	<i>Etheostoma canurum</i>				-/3	-/4
	<i>Etheostoma obeysense</i>	-/4	-/1	-/2	-/1	-/2
	<i>Etheostoma sanguifluum</i>	-/1				

from 0.0 (not detected) to 0.009 mg/liter ( $\bar{x}$  = 0.002  $\pm$  0.003 mg/liter). Levels of Co, Hg, TSS, TVS, and turbidity exhibited substantial variation. Five variables ( $\text{Cl}^-$ , pH, Na,  $\text{SO}_4$ , and Zn) showed considerable variation (standard deviation approaching the mean) in Bear Creek. The levels of Al, Co, Cr, Cu, Hg, Mn, Ni, Zn, TSS, and turbidity exhibited substantial variation in Rock Creek. In addition, Ba,  $\text{Cl}^-$ , Fe, Na,  $\text{SO}_4$ , and TVS had considerable variation in Rock Creek.

Of the 25 variables examined, 13 exhibited significant differences between the two streams. Values for Al, Ca, Mg, Mn, Na, Ni, K, Zn,  $\text{SO}_4$ , pH, conductivity, and hardness were significantly higher ( $P \leq 0.05$ ) and alkalinity was significantly lower ( $P \leq 0.05$ ), in Bear Creek (Figure 3). No significant differences ( $P > 0.05$ ) were observed between sites for Ba, Co, Cr, Cu, Fe, Hg,  $\text{Cl}^-$ , D.O., TSS, TVS, temperature, or turbidity.

#### Assessment of Reclamation Efforts in the Bear Creek Watershed

During fall 1994 Bear Creek macroinvertebrate taxa richness and TNI values approximated the results for spring and fall 1992 and winter 1992, respectively. Macroinvertebrate metric values for this collection were repre-

sentative of a highly tolerant community (mHBI = 7.39) with few intolerant taxa (EPT = 4, %EPT = 20) (Table 1). There were no observed significant differences ( $P > 0.05$ ) between mean values of the fall pre- and post-reclamation samples.

No fish were collected from Bear Creek during 1994 (IBI = 0). In contrast, 358 individuals, representing 5 families and 14 species were collected from Rock Creek (IBI = 34) (Tables 1 and 2). There were no significant differences ( $P > 0.05$ ) observed between the Bear Creek fall pre- and post-reclamation IBI values.

Only three physicochemical variables exhibited significant differences between the pre- and post-reclamation values. Post-reclamation Cu values ( $\bar{x}$  = 0.00 mg/liter) were significantly ( $P \leq 0.05$ ) lower than pre-reclamation values. The post-reclamation values for conductivity ( $\bar{x}$  = 257  $\mu\text{S}/\text{cm}$ ) and K ( $\bar{x}$  = 2.36 mg/liter), however, were significantly ( $P \leq 0.05$ ) higher than the pre-reclamation values.

#### DISCUSSION

The fluctuations observed in taxa richness and TNI from both streams were due in part to the natural phenologies of aquatic macroinvertebrates. The Bear Creek taxa richness and

Table 3. Maximum, minimum, mean and standard deviations of Bear Creek and Rock Creek, McCreary County, Kentucky; physicochemical variables, 1991–1992.

Parameter	Bear Creek				Rock Creek			
	Maximum	Minimum	Mean	Standard deviation	Maximum	Minimum	Mean	Standard deviation
Alkalinity (mg/L)	11.4	0.0	1.66	2.78	18.0	6.33	12.28	4.75
Aluminum (mg/L)	1.78	0.008	0.63	0.57	0.24	0.0	0.06	0.09
Barium (mg/L)	0.064	0.004	0.040	0.020	0.048	0.004	0.025	0.013
Calcium (mg/L)	18.20	5.74	11.38	3.88	6.14	2.87	4.05	0.97
Chloride (mg/L)	6.00	0.0	2.49	2.14	2.40	0.0	1.21	0.84
Chromium (mg/L)	0.009	0.0	0.002	0.003	0.008	0.0	0.002	0.002
Cobalt (mg/L)	0.032	0.0	0.010	0.010	0.006	0.0	0.001	0.003
Conductivity ( $\mu$ S/cm)	280.0	118.0	173.3	48.3	62.0	33.1	44.2	9.4
Copper (mg/L)	0.009	0.0	0.003	0.002	0.018	0.0	0.004	0.006
Dissolved Oxygen (mg/L)	13.1	7.7	10.20	1.6	11.9	8.1	10.1	1.3
Hardness (mg/L)	95.1	22.5	59.2	20.7	25.1	8.4	13.9	5.1
Iron (mg/L)	2.710	0.0	0.34	0.61	0.323	0.004	0.160	0.119
Magnesium (mg/L)	12.30	5.33	8.29	2.26	1.97	0.95	1.15	0.34
Manganese (mg/L)	3.02	0.85	1.82	0.52	0.03	0.0	0.01	0.01
Mercury (mg/L)	0.00020	0.0	0.00004	0.00007	0.00020	0.0	0.00005	0.00008
Nickel (mg/L)	0.057	0.0	0.003	0.01	0.0040	0.0	0.0009	0.0018
pH (SU)	8.2	4.2	5.6	1.2	8.1	6.1	7.0	0.6
Potassium (mg/L)	2.52	0.77	1.54	0.52	0.71	0.43	0.57	0.11
Sodium (mg/L)	7.20	0.0	2.89	2.11	1.43	0.0	0.70	0.47
Sulfate (mg/L)	104.0	30.8	59.6	19.3	13.8	0.0	6.0	4.4
Temperature ( $^{\circ}$ C)	25.0	1.0	10.4	6.3	23.0	1.0	10.6	7.5
Total Suspended Solids (mg/L)	33.0	0.0	5.56	8.35	8.0	0.0	2.4	2.8
Total Volatile Solids (mg/L)	162.0	0.0	28.29	38.64	23.0	0.0	8.83	8.6
Turbidity (NTU)	50.0	0.6	7.8	13.4	8.0	0.8	2.4	2.5
Zinc (mg/L)	0.25	0.03	0.07	0.05	0.065	0.0	0.014	0.021

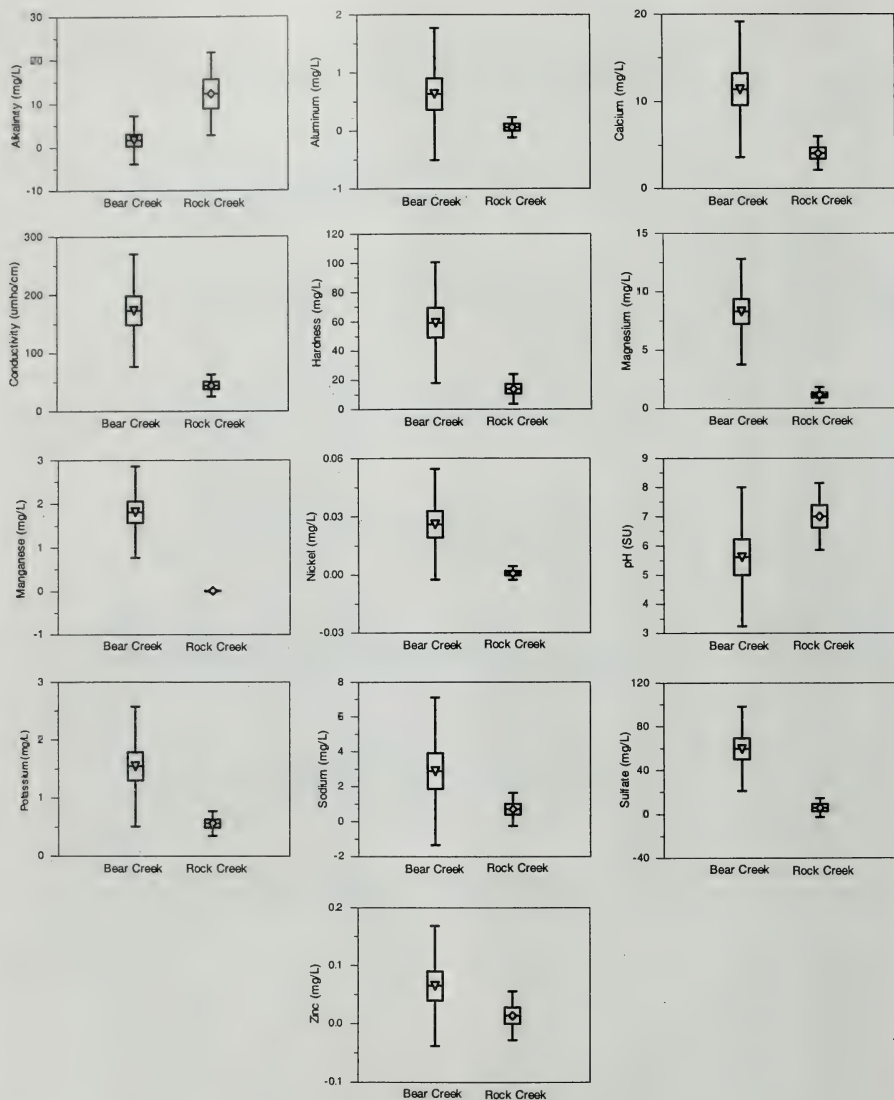


Figure 3. Comparison of significantly different (Mann-Whitney,  $P \leq 0.05$ ) physicochemical variables from Bear Creek and Rock Creek, McCreary County, Kentucky, 1991–1992. Bars on the plots correspond to, in increasing order, mean  $\pm 2 \times$  standard deviation, mean  $\pm 2 \times$  standard error, mean, mean  $\pm 2 \times$  standard error and mean  $\pm 2 \times$  standard deviation.



TNI values, however, were indicative of an impaired system. Low values for these metrics are indicative of AMD-stressed aquatic macroinvertebrate communities (Branson et al. 1984; Cairns et al. 1971). Several genera (14) were collected during summer 1991 from Bear Creek, resulting in the higher taxa richness and TNI values. This included four dipterans (*Ablabesmyia* sp., *Ephydra* sp., *Pentaneura* sp., and *Saetheria* sp.), two odonates (*Neurocordulia* sp. and *Macromia* sp.), one mollusk (*Sphaerium* sp.), one heteropteran (*Rheumatobates* sp.), and one coleopteran (*Oulimnius* sp.) that were collected only from Bear Creek. The highest Rock Creek TNI value was associated with large numbers of the chironomid dipteran *Polypedium* sp. and the heptageniid mayfly *Stenonema vicarium*.

The EPT, %EPT, and %CHIR values for Bear Creek did not necessarily indicate a replacement of intolerant taxa with tolerant taxa; however, they did indicate an impaired system that was not supporting a diverse EPT fauna, the taxonomic groups most affected by AMD (Roback and Richardson 1969). In contrast, Rock Creek supported a diverse EPT fauna. While midges are an integral part of the macroinvertebrate community in most aquatic systems (Coffman and Ferrington 1996), several genera are tolerant of AMD (Armitage and Blackburn 1985; Moon and Lucostic 1979; Roback and Richardson 1969). The AMD-tolerant chironomid genera *Cricotopus* sp., *Procladius* sp., and *Thienemannimyia* gp. were observed in Bear Creek.

Though the validity of using biotic indices originally formulated to assess organic enrichment to evaluate inorganic impacts has been questioned (Johnson et al. 1993), modifications (such as the mHBI used herein) have been proven useful (KDOW 1993; Barbour et al. 1999; García-Criado et al. 1999). The higher Bear Creek mHBI values were due to the presence of several highly tolerant (i.e., tolerance value  $\geq 8.0$ , Lenat 1993) taxa, such as the damselfly *Calopteryx* sp., the worm *Enchytraeus* sp., and beetles *Laccophilus* sp. and *Uvarus* sp. However, lower mHBI values were observed during winter and summer 1991 and fall 1992 (mHBI = 5.46, 4.04 and 4.56, respectively). In contrast, the spring 1991 and 1992 collections from Rock Creek were dominated by the intolerant (i.e., tolerance value

$\leq 2.0$ , Lenat 1993) ephemereiid mayfly *Ephemerella* sp., and exhibited the lowest mHBI values observed from either stream (mHBI = 2.01 and 3.00, respectively).

The  $PS_c$  is a useful, robust measure for comparing the community structure (similarity) of impacted and reference streams. Based on the close proximity of the streams to each other and ecoregional affinities, it would be expected that the  $PS_c$  values would be at least greater than 50%. However, this measure indicated a large dissimilarity between the two streams (Krebs 1989).

Of the four attempts to collect fish in Bear Creek, only one was successful. Obviously this collection had a higher IBI score than the other three collections. However, this rating was based on a single intolerant insectivorous cyprinid (*Cyprinella galactura*) (KDOW 1997), hence the elevated IBI. This should be considered an anomaly, since the pH levels of Bear Creek were consistently outside the range of pH tolerance of minnow (Cyprinidae) species (Heard et al. 1997). This individual was likely either a transient from an unimpacted tributary or an upstream migrant from the South Fork Cumberland River. The loss of fish species can be directly attributed to surface mining in the Bear Creek watershed, as seen in other studies (e.g., Branson et al. 1984). The Rock Creek fish community was very typical of other reference fish communities in the upper Cumberland basin of the Central Appalachians Ecoregion in terms of IBI values and taxa richness (KDOW 1997).

Increases in Al, Ca, Cu, Fe, Mg, Mn, Na, conductivity and  $SO_4$ , and decreases in pH and alkalinity (Becker et al. 1986; Branson et al. 1984; Rikard et al. 1986) were indicative of coal-mining impacts. Total dissolved solids (TDS) can also be used to indicate coal-mining impacts. However, increases in TDS have not been reported for watersheds in which mining has not occurred for some time (Becker et al. 1986), such as the Bear Creek watershed.

The pH level of a stream is commonly believed to be the most reliable indicator of AMD. However, the presence of carbonates and alkaline soils can raise alkalinity in some streams, and pH levels are therefore usually more buffered and do not reflect the actual level of impact in the system (Moon and Lu-

costic 1979; Rikard et al. 1986). Consequently,  $\text{SO}_4$  and Mn are more reliable indicators of AMD (Branson et al. 1984; Rikard et al. 1986), and Bear Creek  $\text{SO}_4$  and Mn values were significantly higher than for the un-impacted Rock Creek.

While Stucki (1995) documented some slight improvements in water quality in the larger (third-order) streams in the Tennessee portion of the watershed following reclamation, no improvements were observed at the mouth of Bear Creek. However, reclamation has occurred at only 3 of the 33 identified sites in the Bear Creek watershed. Targeting more BMPs in the critical areas in the watershed may result in more demonstrable improvements in water quality (Spooner and Line 1993).

### CONCLUSIONS

Attempts are still being made by several state and federal agencies to abate AMD in the Bear Creek watershed. For example, approval was granted for reclamation efforts under the Watershed Protection and Flood Prevention Act (P.L. 83-566) (USDA 1997). It has been estimated, however, that the average cost for remediation in the watershed is ca. \$10,700 per ha (USDA 1991). Newer restoration techniques, such as the direct application of limestone to AMD-impacted streams (e.g., Clayton et al. 1998; Clayton and Menendez 1996), may prove to be both ecologically beneficial and cost-effective. However, more research is needed to assess the effectiveness and possible ecological side effects (i.e., increased total settleable solids) of these techniques.

Because of the severity and duration of the anthropogenic impacts to Bear Creek, significant water quality improvements may not occur for several years following remediation (Cairns et al. 1971; Wallace 1990). For example, Pond (2000) found that differences still existed between the macroinvertebrate communities of two first-order streams with different disturbance histories. One of the streams had been impacted by mining over 25 years ago, but no disturbance had occurred since that time. The timeframe for recovery of both the macroinvertebrate and fish communities is ultimately dependent upon the availability of suitable habitat and the proximity of

colony pools once the AMD impacts have been ameliorated (Cairns et al. 1971; Nelson and Roline 1996).

Future monitoring efforts should take the following into consideration. The macroinvertebrate and fish methodologies used in this study (i.e., Surber sampler and seine) are not effective in the boulder-dominated substrate of the study streams (McMurray pers. obs.); it is recommended that future monitoring employ kick-nets and a backpack electrofisher. Also, additional data on the ichthyofauna of upper Cumberland River basin streams in the Central Appalachian Ecoregion are necessary for the IBI to be effectively used as an indicator of water quality there. Future physicochemical monitoring should be conducted on a regular basis, regardless of flow conditions, to ascertain the success of any remediation efforts. Finally, at least 2 to 3 yrs of both pre- and post-restoration biological and physicochemical data should be collected to increase statistical sensitivity (Spooner et al. 1990; USEPA 1997).

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Appendix A. Macroinvertebrates collected from Bear Creek (BC) and Rock Creek (RC), McCreary County, Kentucky, 1991–1992. Values are presented as BC/RC, during winter [win], spring [spr], summer [sum] and fall. A—indicates the organism was not collected.

	Taxa	1991				1992			
		Win	Spr	Sum	Fall	Win	Spr	Sum	Fall
Enchytraeidae	<i>Enchytraeus</i> sp.	3/4	2/-	37/-	7/-	4/-	6/-	4/-	3/-
Lumbriculidae	<i>Lumbriculus</i> sp.	-/4	-/1	-/1	1/8	-/1	-/3	-/2	-/3
Naididae				-/1					
Tubificidae				-/3					
Ancyliidae	<i>Ferrissia</i> sp.			-/1					
Physidae	<i>Physella</i> sp.			-/14	-/2	-/1			
Pleuroceridae	<i>Elintia</i> sp.				-/8				
Planorbidae	<i>Helisoma</i> sp.	-/30		-/7					
Sphaeriidae	<i>Sphaerium</i> sp.			1/-					
Crangonyctidae	<i>Crangonyx</i> sp.					1/-			
Cambaridae	<i>Cambarus</i>								
	<i>cumberlandensis</i>		-/1		-/2				-/1
	<i>Cambarus distans</i>		2/2	2/1	4/12		4/3	-/1	4/4
	<i>Orconectes placidus</i>		-/1	2/4	1/-	1/-	-/3		1/-
Baetidae			-/18	-/6	-/6	-/7		-/21	-/8
	<i>Acentrella</i> sp.			-/13	-/3			-/13	-/11
	<i>Baetis</i> sp.				-/3		-/2	-/1	-/2
	<i>Centropilum</i> sp.				-/4	-/6		-/1	-/1
Baetiscidae	<i>Baetisca</i> sp.								
Caenidae	<i>Brachycerus</i> sp.	-/1		-/1					
	<i>Cacius</i> sp.								
	<i>Cercobranchys</i> sp.		-/99	-/17	1/2	-/1	2/69		-/10
Ephemeriidae	<i>Ephemerella</i> sp.	-/26		1/-	-/15	-/37	-/3		1/-
	<i>Eurylophella</i> sp.	-/2			-/4	-/1	3/-		
	<i>Hexagenia</i> sp.		-/2				-/5		
	<i>Ephemera</i> sp.			-/74			-/16		-/1
	<i>Cinygmula subaqualis</i>	-/1	-/47	-/6	-/6	-/19	-/39	-/7	-/19
Ephemeriidae	<i>Epeorus</i> sp.	-/8	2/32	-/1					
Heptageniidae	<i>Heptagenia</i> sp.	-/1		-/8					
	<i>Leucocrota</i> sp.				-/13			-/2	
	<i>Stenonema</i> sp.	-/9	-/9	-/36	-/221	-/15	-/25	-/30	
	<i>Stenonema femoratum</i>		-/14		-/15	-/12	-/1		
	<i>Stenacron</i>								
	<i>interpunctatum</i>		-/1		4/1		-/12		
	<i>Stenonema</i>			-/4					
	<i>medtopunctatum</i>		-/2	-/127	-/80		-/18	-/15	-/109
	<i>Stenonema vicarium</i>	-/2							





Appendix A. Continued.

	Taxa	1991				1992			
		Win	Spr	Sum	Fall	Win	Spr	Sum	Fall
Belostomatidae	<i>Belostoma</i> sp.				1/-				
Corixidae	<i>Sigara</i> sp.		1/-	1/-	1/-				
Gerridae	<i>Limnoporus</i> sp.		1/-	1/-	-1				
	<i>Rheumatobates</i> sp.			1/-					
	<i>Trepobates</i> sp.			-2					
Velidae	<i>Microvelia</i> sp.			-3	2/3		-1		-4
	<i>Rhagovelia</i> sp.			-1	-2				-9
Corydalidae	<i>Corydalus cornutus</i>	-4	-3	-6	-22	1/-	-3		
	<i>Nigronia</i> sp.				3/-				
	<i>Nigronia serricornis</i>			2/-		1/-	-1	-1	
Sialidae	<i>Sitalis</i> sp.			7/-	-1				
Brachycentridae	<i>Micrasema</i> sp.			-7					
Calamoceratidae	<i>Heteroplectron americanum</i>								
Glossosomatidae	<i>Glossosoma</i> sp.	-2	-3	-1		-2	-1	-2	-9
Goeridae	<i>Goera</i> sp.	-1							
Helicopsychidae	<i>Helicopsyche borealis</i>			-2		-11	-3		
Hydropsychidae	<i>Ceratopsyche sparna</i>	-5	-5	-4	-5			1/-	11/15
	<i>Ceratopsyche ventura</i>		-3				-2	-11	
	<i>Cheumatopsyche</i> sp.	-4			2/16	-2	1/10	1/-	-7
	<i>Diplectrona modesta</i>		-8	5/-	1/9	1/-	-1		6/1
	<i>Hydropsyche betteni</i>					3/-			1/-
	<i>Lepidostoma</i> sp.		-3		2/-				
Lepidostomidae	<i>Oecetis</i> sp.		1/-	-3	1/-				
Leptoceridae	<i>Pycnopsyche</i> sp.		-1	-1		-2	2/-		3/-
Limnephilidae	<i>Chimarra</i> sp.	1/-	-2	-10	-156	-1	-7	-5	-16
Philopotamidae	<i>Dolophiles distinctus</i>	-8			-1				-2
	<i>Wormidia</i> sp.				-1				-3
Polycentropodidae	<i>Ceratonia</i> sp.					-1			1/-
	<i>Nyctiophylax</i> sp.								
	<i>Polycentropus</i> sp.		-2	-1	1/7	1/1	-1		-1
Rhyacophilidae	<i>Rhyacophila</i> sp.	-1			-6				
Circulionidae		1/-			3/-				
Dryopidae	<i>Helictus</i> sp.		2/-						1/-
	<i>Helictus basalis</i>	-1	2/3	-3	1/2		-1	-6	

Appendix A. Continued.

Taxa	1991				1992			
	Win	Sp	Sum	Fall	Win	Sp	Sum	Fall
<b>Dytiscidae</b>								
<i>Dytiscus</i> sp.			11/-					
<i>Hydroporus</i> sp.			8/7			2/2		
<i>Laccophilus</i> sp.		2/-	1/-	7/-		-1	-1	
<i>Neoporus</i> sp.		2/-		2/1				
<i>Uvarus</i> sp.						-2		
<b>Elmidae</b>								
<i>Macronychus glabratus</i>			-6					-1
<i>Dubiraphia</i> sp.			-1			-3	-7	-2/8
<i>Optoserius</i> sp.		-1	-2/29				-2	
<i>Oulimnius laticutulus</i>			2/-	-7	-6			-1/1
<i>Promoresia</i> sp.				-9				-1
<i>Stenelmis</i> sp.	1/-	-1	-2			-2		-1
<i>Sperchopsus</i> sp.						-1		-1
<i>Tropisternus</i> sp.		-3	2/5	-7	1/3	1/6		-4
<i>Psaphenus herricki</i>	-2		4/2	-2	-1	-5	-1	-6
<i>Atherix variegata</i>				-3	3/-	-2		
<b>Hydrophilidae</b>								
<i>Abdusomyia</i> sp.			2/-		-2			-6
<i>Brillia</i> sp.			-8	-8/5			-1	
<i>Cladotanytarsus</i> sp.			-35	-6			-1	
<i>Corynoneura</i> sp.			1/-	1/2	-1			-8
<i>Cricotopus</i> sp.	-11		8/16	2/-	3/8	2/-	3/-	-1
<i>Cryptochironomus</i> sp.			2/17					
<i>Endochironomus</i> sp.						-2		
<i>Eukiefferiella</i> sp.			-1					
<i>Glyptotendipes</i> sp.		-3	-10	1/4	-4	-18	-19	
<i>Micropectra</i> sp.			2/6	1/4		1/-	9/2	-6
<i>Microtendipes</i> sp.			-1					
<i>Nanocladius</i> sp.			-2					
<i>Omisus</i> sp.		-13				-1	-1	
<b>Orthocladinae</b>								
<i>Parakiefferiella</i> sp.	-24		-5					
<i>Parametrioctenium</i> sp.	-55		-4	-1		-8		
<i>Pentaneura</i> sp.			1/-					
<i>Phaenopsectra</i> sp.	5/-	1/-	45/82	22/-	5/2	3/2	7/-	1/-
<i>Polypedilum</i> sp.		-6	2/169	-34		-9	1/7	-16
<i>Procladius</i> sp.			-1	-3		-13		
<i>Rheocricotopus</i> sp.			-1		-3	-1		

Appendix A. Continued.

	Taxa	1991				1992			
		Win	Spr	Sum	Fall	Win	Spr	Sum	Fall
	<i>Rheotanytarsus</i> sp.	-/18		-/19	-/4	1/1	-/1		
	<i>Saetheria</i> sp.			3/-					
	<i>Stempellina</i> sp.				-/8				
	<i>Stempellinella</i> sp.								-/1
	<i>Stenochironomus</i> sp.			-/12	1/2				
	<i>Tanytarsus</i> sp.	-/12	1/-	7/29	-/1	-/1			
	<i>Thienemannella</i> sp.	1/-		3/-					
	<i>Thienemannimyia</i> gr.	-/6		-/11		1/-			
	<i>Zaurelmigia</i> sp.	1/-		-/1		3/-			
	<i>Hemerodromia</i> sp.						-/1		
	<i>Ephydra</i> sp.			1/-					
Empididae									
Ephyridae									
Simuliidae									
	<i>Prosimulium</i> sp.	-/416	-/3		-/3	-/69	-/23		
	<i>Simulium</i> sp.			-/10	-/40	1/43	1/6	-/98	-/31
Tabanidae	<i>Chrysops</i> sp.			1/-	1/-		1/1	-/1	
Tanyderidae	<i>Protoplasa fitchii</i>			-/2			-/2	-/1	
Tipulidae	<i>Antocha</i> sp.	-/1			-/3		-/2	-/2	
	<i>Dicranota</i> sp.		-/1				-/3		
	<i>Hexatoma</i> sp.	1/-	-/3	-/1	-/6			1/-	-/1
	<i>Limnophila</i> sp.	2/4		2/1	1/-	1/1	-/9	1/-	2/3
	<i>Tipulasp.</i>			1/-	2/6	2/-			



## NOTES

**Central nervous system disorder in a woodchuck (*Marmota monax*) in central Kentucky caused by a nematode in the genus *Baylisascaris*.**—In central Kentucky in June 2000 a young adult female woodchuck was observed with unusual clinical signs suggesting a central nervous system disorder. The animal was found during the day in a pasture on a road leading to a barn previously used to stable horses. Several wild animals, including woodchucks, raccoons, and skunks, were using the barn for shelter. Since the animals were causing considerable damage to the foundation and ground in the barn, the animals, primarily woodchucks, were captured with live traps and moved to another location on the farm. The owner was instructed to report any additional animals showing unusual clinical signs. No additional reports were received.

The affected woodchuck was very uncoordinated and unable to walk or run normally. When it attempted to run, it would stumble and roll over on its side and back. It did not appear to be mean or aggressive. The woodchuck was captured alive and taken to a veterinary clinician, and a tentative diagnosis of rabies was made. The animal was euthanized and a postmortem was performed at the Livestock Disease Diagnostic Center, University of Kentucky.

The carcass was received shortly after death and was well preserved. There were no gross lesions in the brain or spinal cord. One-half of the brain was removed for rabies examination. This tissue was negative for rabies. The surface of the lungs and liver contained multiple granulomas 1 to 2 mm in diameter. The serosal surface of the stomach, small intestine, cecum, and colon also contained granulomas. However, unlike in other organs, the granulomas were so extensive that there was very little normal tissue between them.

Samples of the brain, lung, liver, and gastrointestinal tract were taken for parasitologic examinations. Attempts were made to recover nematodes by cutting up the various tissues into small pieces; these were then placed in a separate funnel of the Baermann apparatus. Larval nematodes, measuring 1.078 to 1.323 mm long, were found in the brain, lung, and colon. Some larvae in tissue of the large intestine, kept at 4.5° C, were still alive 11 days after death of the woodchuck.

Samples of all organs were fixed in 10% neutral-buffered formalin and processed for histopathologic examination. Sections were cut at 6  $\mu$  and stained with hematoxylin and eosin. The brain and spinal cord contained multiple granulomas, each with nematode larvae varying slightly in size. The larvae were surrounded by mixed inflammatory cells, including eosinophils, neutrophils, mixed mononuclear cells, and a thin capsule made up of fibroblasts. Similar granulomas occurred in the lungs, heart, liver, kidneys, and gastrointestinal tract (mostly in the colon). The granulomas, containing nematode larvae, were present in all layers of the colon but were most extensive

in the muscularis mucosa and near the serosal surface (Figures 1 and 2).

The larval nematodes were identified as *Baylisascaris* sp. by Dr. Kevin Kazacos, Purdue University, West Lafayette, Indiana, the world authority on these parasites. It is difficult to identify to species the larval stages of this genus. Characteristics and general information on *Baylisascaris* sp., mentioned in this paper, are mostly from Kazacos (1, 2, 3, 4).

There are at least eight species of *Baylisascaris*; seven of these mature in carnivores. These parasites may use a variety of intermediate hosts, including many species of mammals and birds; all become infected by ingesting infective eggs from the environment. In these hosts, the parasites produce a condition called larva migrans. The most frequent clinical problem caused by the larval stages is a central nervous system disorder brought about by migration of the larvae in the brain and spinal cord. This disorder may be mistaken for rabies as in this case. The most common species of *Baylisascaris* causing larva migrans is *B. procyonis* from raccoons; a few cases have been reported for *B. columnaris* in skunks. Eggs of these ascarids, or roundworms, are passed in the feces of the definitive host (raccoons or skunks) (4). Embryonation to the infective stage occurs in the eggs in as few as 11–14 days. Eggs can remain viable for long periods (months to years). When eggs containing infective larvae are accidentally ingested by young raccoons and skunks, development to the adult stage is completed in the small intestine. Usually, no problems due to the infections are evident in these hosts. However, if the eggs are swallowed by other hosts like woodchucks or rabbits, which serve as intermediate hosts for the raccoon and skunk roundworms, larvae do not mature. They penetrate the intestine and migrate to various areas of the body, including the brain, spinal cord, eyes, intestines, lungs, liver, and muscle. Larvae in the central nervous system may cause damage so severe that the host dies. Injury to the eye may result from larvae entering that site.

*Baylisascaris* spp. can be a problem in both wild and domestic animals. To date, over 90 species of mammals and birds have been killed by *B. procyonis* in North America (4). Humans also can be infected with these larvae, which may cause not only eye problems but also death from central nervous system damage. Over a dozen cases of neurologic disease are known, mostly involving young children less than 2 years old (4). Caution should be observed in areas where raccoons and skunks defecate and deposit ascarid eggs. Typical residences for raccoons are barn floors and lofts where there can be build up of feces (latrines). Raccoon latrines also occur on fallen logs, stumps, tree bases, and roofs and decks. Where possible, raccoons and skunks should be prevented or discouraged from locating in areas inhabited or frequented by humans. Removal of raccoon feces, such as from barn lofts or decks, should be done periodically if these animals cannot

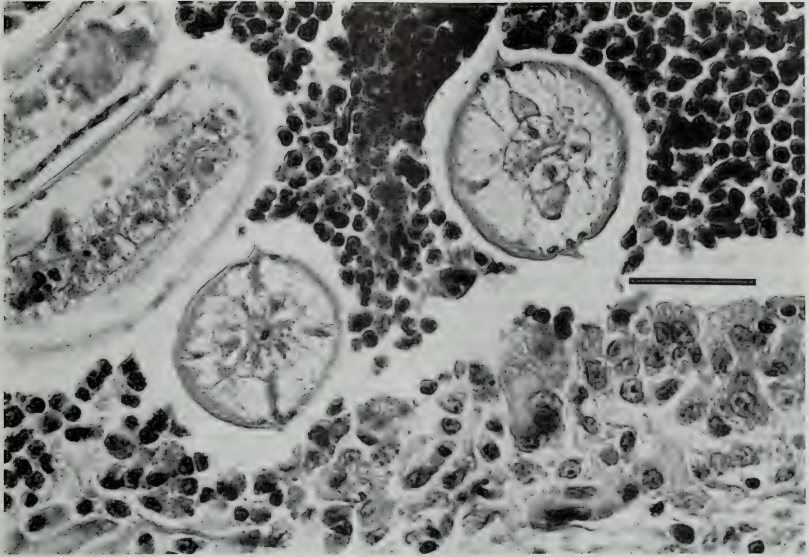


Figure 1. Histologic section of wall of colon of woodchuck showing portions of larval *Baylisascaris* sp. (pointed structures are cervical alae). Scale bar = 30  $\mu$ m.

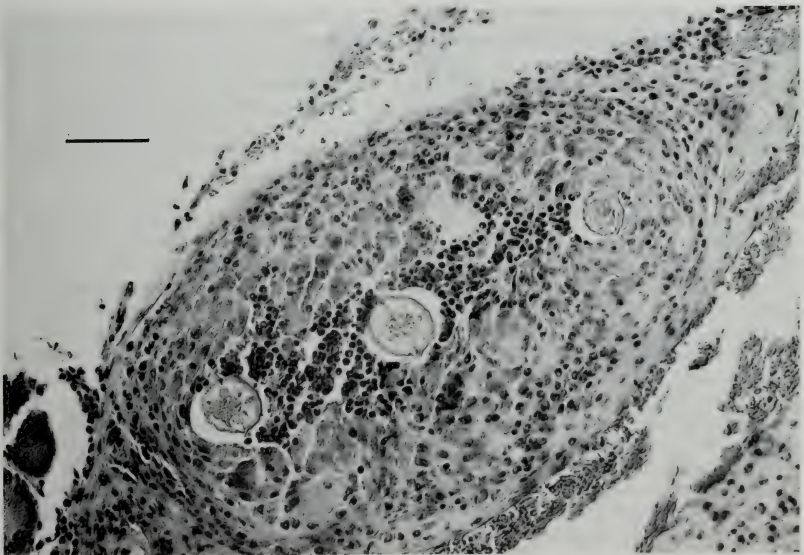


Figure 2. Histologic section of wall of colon of woodchuck showing portions of larval *Baylisascaris* sp. (pointed structures are cervical alae). Similar to Figure 1 but at lower power. Scale bar = 50  $\mu$ m.



be kept from living there. When doing this, a person should wear a face mask and disposable gloves to lessen the chance of being infected with embryonated eggs. Removed contaminated material should be destroyed by burning; boiling water can be poured on contaminated decks. Young children should be kept away from contaminated areas; the keeping of raccoons or skunks as pets should be discouraged. Detailed question/answer information on *Baylisascaris* infections is included in two publications by Kazacos (1, 3); an extensive review is also available (4).

This research paper (No. 01-14-36) was done in connection with a project of the Kentucky Agricultural Experiment Station and is published with the approval of the director.

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**Rediscovery of the Ashy Darter, *Etheostoma cinereum* (Pisces: Percidae) in Buck Creek, Pulaski County, Kentucky.**—*Etheostoma cinereum* (Storer) (ashy darter), the only species in the subgenus *Allohistium*, is endemic to the Cumberland and Tennessee River drainages in Alabama, Georgia, Kentucky, Tennessee, and Virginia. It is thought to be extirpated from Alabama and Georgia (1) and has not been collected in Virginia in over 30 years (2). However, populations persist in the Highland Rim and Cumberland Plateau physiographic provinces in Kentucky and in the Highland Rim and Ridge and Valley provinces in Tennessee. *Etheostoma cinereum* is listed as threatened in Tennessee (1) and has been downgraded from threatened to special concern in Kentucky (3, 4). The ashy darter occurs in clear, small to medium-sized upland streams. It often is associated with some form of cover such as boulders, bedrock slabs, vegetation (e.g., *Justicia*), and tree snags, and over substrates of bedrock and gravel with minimal silt deposits (1, 5). The largest known population exists in the Little River, Blount County, Tennessee, where a particular pool was estimated to have contained 300 individuals (1). The ashy darter has a sparse distribution in Kentucky and has been reported only from six streams: Horseshoe Creek, Jackson County (6); Red River, Logan County; Rockcastle River, Rockcastle County; Buck Creek, Pulaski County; Little South Fork of the Cumber-

land River, Wayne County; and South Fork of the Cumberland River, McCreary County (7). According to Burr and Warren (5), the Little South Fork of the Cumberland River and the Rockcastle River apparently have the largest populations in Kentucky, with individuals occasionally being common in some locations. The ashy darter was thought to be extirpated from Buck Creek and from the Red River, as the species had not been collected from either watershed since 1955 and 1957, respectively (7, 8).

Fish were sampled at 10 sites on Buck Creek using a 3.4 m × 1.2 m, 0.3-cm-mesh seine. Specimens were preserved in a 10% formaldehyde solution, transferred to the laboratory for identification and permanent storage in 70% ethanol, and identified using Etnier and Starnes (1) and Page and Burr (9). Collections are deposited in the Branley A. Branson Museum of Zoology at Eastern Kentucky University.

On 24 May 1996 two specimens of *Etheostoma cinereum* were collected from Buck Creek at a site located just downstream from the confluence with Lick Branch (37°14'49.2"N, 84°32'38.9"W), ca. 3.2 km SW of Woodstock and 4.8 km SSE of Clarence, Pulaski County, Kentucky. The stream drains ca. 221 km<sup>2</sup> and is fourth order at this site. A large cobble-and-gravel riffle was bordered both upstream and downstream by long, deep pools over silt and sand. The fish were collected 4 m downstream of the riffle in a section of pool ca. 0.8 m in depth, along the right downstream side and in sluggish to slow current. The substrate in the immediate area was dominated by sand and silt, with some gravel present. On 7 Sep 1996 two additional specimens were collected using a backpack electro-shocker. These specimens were collected in the same immediate area, identified, and then released. Other sites along Buck Creek (10 sites) yielded no *E. cinereum*.

This collection of *E. cinereum* is the first reported in over 40 years in Buck Creek, and represents the most upstream location. All four of the collected specimens were young of the year, indicating a viable population. Historical collections have been made from Buck Creek at the old KY 80 bridge at Stab, Pulaski County (27 Jul 1954, J. R. Charles; 14 Sep 1955, C.R. Gilbert and B.C. Franklin), and at the confluence with Brushy Creek, Pulaski County (14 Sep 1955, C.R. Gilbert and B.C. Franklin) (8). The site reported herein represents a ca. 7.5 km upstream from previous localities in the Buck Creek system. Cicerello and Butler (8) suggested that populations may still exist downstream from the historical sites between the old KY 80 bridge and the KY 192 bridge. Few collections have been made in this reach, with the KY 1003 bridge being the only consistently sampled site. More intensive searches within this reach are needed.

Protection of this and other populations of *E. cinereum* is limited since Kentucky's endangered or threatened species designations are without regulatory force. This population may be in jeopardy due to continuing agricultural practices, dredging, and proposed channelization of selected stream reaches in Buck Creek.

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V. Bishop, J. Secrest-Board, T. Oliver, C. Kirk, D. Peake, and C. Peters for help in the field; P. Ceas for species verification; G.A. Schuster, S. McMurray, and two anonymous reviewers for their review of the paper and helpful comments; G.A. Schuster and the Department of Biological Sciences, Eastern Kentucky University, for initiating the study on Buck Creek, which led to this rediscovery of *E. cinereum*.

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**Rare and Extirpated Biota of Kentucky: 2001 Update.**—The Kentucky State Nature Preserves Commission (KSNPC) published a revised list of rare and extirpated biota of Kentucky in 2000 (1). That list was based on distributional and ecological data available as of 1 Jan 2000. With the assistance of many scientific authorities, we are continually learning more about the flora and fauna of the state. As part of an ongoing data management process, KSNPC conducts annual reviews of the nomenclature and statuses of the state's rare biota. Herein we provide an update based on data available through 30 Apr 2001. Over time, the evolution of KSNPC's list of rare biota has reflected in part the acquisition of knowledge of less well-known groups of plants and animals. In this vein, most of the 2001 update represents the inclusion for the first time of several groups of invertebrate animals that inhabit cave ecosystems.

Table 1. Conservation status changes for rare and extirpated Kentucky biota, 2001.

	KSNPC Status <sup>1</sup>	
	Old	New
Insects		
<i>Pyrgus wyandot</i>		
Appalachian grizzled skipper	T	H

<sup>1</sup> T = Threatened, H = Historic.

The methods and status categories used herein follow KSNPC (1). Species whose conservation statuses are being changed are given in Table 1; changes in nomenclature and additions to the list are presented in Tables 2 and 3, respectively. The source for plant names in this update is Kartesz (2). Sources for animal names are as follows: arachnids—Harker and Barr (3), Malcolm and Chamberlin (4), Muchmore (5, 6, 7); diplopods—Shear (8); and insects—Arnett (9), Christiansen and Bellinger (10), McCafferty (11), Schweitzer (12), and Stewart and Stark (13). We welcome questions or comments about this update or KSNPC (1).

We thank the following individuals for sharing information and for their assistance: Nicole Capuano, The Association for Biodiversity Information; Kenneth A. Christiansen, Grinnell College; Charles V. Covell Jr., University of Louisville; David C. Culver, American University; Lorran D. Gibson; Scott A. Grubbs, Western Kentucky University; Ronald L. Jones, Eastern Kentucky University; Boris C. Kondratieff, Colorado State University; Jerry J. Lewis; Pam Lyons, Kentucky Department For Libraries and Archives; and Greg J. Pond, Kentucky Division of Water.

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Table 2. Nomenclature changes for rare and extirpated Kentucky biota, 2001.

Old name	New name
Plants	
<i>Arabis hirsuta</i> var. <i>adpressipilis</i>	<i>Arabis hirsuta</i> var. <i>adpressipilis</i>
Hairy rock-cress	Hairy rock-cress
<i>Corydalis sempervirens</i>	<i>Corydalis sempervirens</i>
Pale corydalis	Rock harlequin
<i>Trillium pusillum</i> var. <i>pusillum</i>	<i>Trillium pusillum</i>
Least trillium	Least trillium
<i>Trillium pusillum</i> var. <i>ozarkanum</i>	<i>Trillium pusillum</i>
Ozark least trillium	Least trillium

Table 3. Additions to the list of rare and extirpated biota of Kentucky, 2001.

	KSNPC <sup>1</sup>	US
<b>Plants</b>		
<i>Hydrocotyle ranunculoides</i>		
Floating pennywort	E	—
<i>Stenanthium gramineum</i>		
Eastern featherbells	T	—
<i>Vaccinium erythrocarpum</i>		
Southern mountain cranberry	E	—
<i>Viburnum lantanoides</i>		
Alderleaf viburnum	E	—
<b>Arachnids</b>		
<i>Herperonemastoma inops</i>		
A cave harvestman	S	—
<i>Kleptochthonius cerberus</i>		
A cave pseudoscorpion	S	—
<i>Kleptochthonius ereticus</i>		
A cave pseudoscorpion	T	—
<i>Kleptochthonius hageni</i>		
A cave pseudoscorpion	S	—
<i>Kleptochthonius hubrichti</i>		
A cave pseudoscorpion	T	—
<i>Kleptochthonius microphthalmus</i>		
A cave pseudoscorpion	T	—
<i>Tyrannochthonius hypogaeus</i>		
A cave pseudoscorpion	S	—
<b>Diplopods</b>		
<i>Pseudotremia amphiorax</i>		
A cave millipede	T	—
<i>Pseudotremia carterensis</i>		
A cave millipede	S	—
<i>Pseudotremia merops</i>		
A cave millipede	T	—
<i>Pseudotremia spira</i>		
A cave millipede	T	—
<i>Pseudotremia unca</i>		
A cave millipede	T	—
<b>Insects</b>		
<i>Acroneturia kosztarabi</i>		
A perlid stonefly	S	—
<i>Allocapnia cunninghami</i>		
A capniid stonefly	II	—
<i>Habrophlebiodes celeteria</i>		
A leptophlebiid mayfly	II	—
<i>Hansonoperla hokolesqua</i>		
A perlid stonefly	S	—
<i>Nixe flowersi</i>		
A heptageniid mayfly	II	—
<i>Pseudanophthalmus cnephosus</i>		
A cave beetle	T	—
<i>Pseudosinella espanita</i>		
A cave springtail	S	—
<i>Timpanoga provanshai</i>		
An ephemeroellid mayfly	II	—

S = Special Concern, T = Threatened, E = Endangered, II = Historic.

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**Abstracts of Some Papers Presented at the  
2000 Meeting of the  
Kentucky Junior Academy of Science  
Edited by Robert J. Barney**

**BEHAVIORAL & SOCIAL SCIENCES**

Liberal or conservative? How political views are divided. CHRISTOPHER YEE, duPont Manual High School, Louisville, KY 40208.

The labels of "liberal" and "conservative" have long been used to indicate, whether positively or negatively, the political standing of an individual or group. Those under the labels supposedly stand for different things; however, more people today seem to support the standings of either side. This study was conducted to test the degree of difference between the views of liberals and the views of conservatives. It was hypothesized that liberal and conservative views would tend to overlap between those who consider themselves to be one or the other. To test this hypothesis, a survey was created to gather information on the political position of a group of people. The questions were made based on knowledge of politically and socially sensitive issues, and the participants answers would determine how liberal or conservative they were. The questions were each given a point value, and each had a liberal response and a conservative response. A more negative point value would be more liberal, while a more positive value would be more conservative. Judging from the results of the survey, the hypothesis is supported. Self-proclaimed liberals and conservatives both agreed with the majority of the views that they should have, although they also agreed with many stances held by the opposite side. On average, the answers of both liberals and conservatives reflected an opposite view more than 25% of the time.

Stereotype threat among female high school students: does it impact performance in mathematics? NEVILLE G. RYANT, duPont Manual High School, Louisville, KY 40208.

Are female high school students involved in studies of mathematics and the sciences vulnerable to stereotype threat? To answer this question, participants were placed under either a neutral, non-threatening or a stereotypically threatening condition before being asked to take a mathematics portion of the GRE. Additionally, participants performed a word completion task in order to determine whether cognitive activation of stereotypical constructs had, indeed, occurred. To participants in the non-threatening condition, six neutral commercials were shown; participants under the threatening condition were shown four neutral commercials and two commercials containing negative stereotypes of women. The experiment found no evidence of a significant negative impact on performance in either males or females when exposed to the stereotype-containing commercials. However, results of the word completion task indicated that cognitive activation of stereotypical constructs occurred when par-

ticipants were exposed to the stereotypical commercials. Thus, it was concluded that while the threatening condition activated stereotypical constructs, female participants did not experience the condition of stereotype threat. The lack of confirmation of stereotype threat was attributed in part to the variability inherent to such an exceedingly small sample size. As well, it was determined that the difficulty of the mathematics section used may not have been great enough to place participants in a position where they were at risk of confirming negative stereotypes.

Internet addiction: a survey of high school students. ROBERT BERN, duPont Manual High School, Louisville, KY 40208.

The Internet is a new form of communication that is receiving a lot of attention through the media. The Internet is highly beneficial in research and resources. However, recent studies have shown that the Internet can also be addictive. A group of volunteer students were asked to fill out a survey that asked questions regarding the Internet. The first half of the survey collected demographic information such as age, magnet program, and experience level. The second half of the survey asked 20 Internet-usage-related questions, with answers ranging one to five based on an answer key. The answers on the second half were totaled and compared to the level of risk chart. According to the chart, 13 of the 139 students surveyed were at the Warning level and none was in the High Risk level (the rest were in the Low Risk level). The data were also analyzed based on demographic information. It was found that the female average score (30.965) was higher than the male average score (29.018). The data also support that the students in the Communication/Media Arts and Visual Arts magnets had higher scores (33.5 and 31.4, respectively) than the other magnets: Math/Science/Technology (30.4), High School University (29.7), Youth Performing Arts School (28.2).

The effect of multilingualism on the learning of a new language. NGOCUYEN V. NGUYEN, duPont Manual High School, Louisville, KY 40208.

Long-term memory and short-term memory are essential to learning. Learning of anything, including languages, would be impossible without the existence of memory. The chief center for language is located on the left side of the brain. Multilingualism is the ability to read, write, speak fluently, and understand more than one language. To achieve this, a person does not necessarily have to learn both languages at birth. The purpose of this experiment is to test the kind of effect multilingualism would have on the progress of a person learning a new language. The hypothesis that multilingual (bilingual) individuals would



do better than monolingual individuals in the progress of acquiring a foreign language were not supported. Subjects at Advance French 11 level were asked to complete a written examination. Scores from each test will reveal the individual competency in the language. Even though the results showed that the average score for bilinguals were higher than the average score for monolinguals, the result did not support the hypothesis completely. The null hypothesis was not rejected, because the t-test showed that the differences were insignificant.

Racial awareness between generations. JENNIFER E. GLASS, duPont Manual High School, Louisville, KY 40208.

This project was completed in hopes of finding a common theme between generations on the subject of racial awareness. There has always been racism in America. Slaves got their freedom in 1865 with the end of the Civil War, and segregation was ended in 1954 by Brown vs. Board of Education, but even after these attempts at equality, many Americans believe that racism is still present in the United States. The hope is that each generation is becoming less racist than the last. Many people believe that the way to do this is to teach the younger generation not to notice race, that everyone is the same. This experiment involved 75 human subjects, 25 from each of three age groups. The subjects were asked to describe several pictures, some of which were humans of different races. The responses were recorded on a survey form where I could distinguish between those who mentioned race and those who did not. The data were compiled and conclusions were drawn.

The effect of a fragrance on short-term memory in senior citizens. SHANEN FUCHS, duPont Manual High School, Louisville, KY 40208.

The sense of smell, or olfaction, is used when perceiving substances through the stimulation of nerves in the nasal cavity by particles given off by that substance. Some olfactory areas of the brain are directly connected to the limbic system, which is responsible for evoking emotions and forming memories. Could a fragrance help a person to remember more information than that person normally would? Ten senior citizens ages 70 to 92 were gathered in a room that had no smell and were asked to look at a list of 15 food items for 30 seconds. The subjects were then asked to recall as many items as they could. Another group of 10 senior citizens in the same age range was placed in a room that was scented with an apple fragrance and was asked to look over the same list of 15 words. This group was also asked to recall as many items as possible and the results were recorded. The data showed that the people in the room with the fragrance remembered an average of only 0.5 more items than those who were in the room with no fragrance remembered. Because of such a small difference, the t-test results showed that the difference was insignificant and, therefore, that the hypothesis is not supported by the data.

## BIOLOGICAL TOPICS

Effect of dopamine depleting agent tetrabenazine on malonate-induced lesion. MINDY SIMONS, Nicholas County High School, Carlisle, KY 40311-9124.

Huntington's Disease is a fatal autosomal, dominantly inherited, neurodegenerative disorder affecting ca. 5-10 per 100,000 people. In 1993, a mutated gene on chromosome 4 consisting of an expanded number of CAG trinucleotide repeats was discovered. Neuropathologically HD causes a progression of neurodegeneration in the corpus striatum that appears to be a type of cell death called apoptosis. One factor that appears to be related to this cell death is the sensitivity of the striatum to the presence of dopamine. Dopamine in high concentrations is neurotoxic. Moreover, removal of striatal dopamine has been shown to confer protection in several models of cell death. To evaluate this finding a suitable model is required. Intra-striatal injections of malonate, a mitochondria inhibitor, produces lesions in rats that mimic the neuropathology of HD. In previous studies dopamine levels were reduced by surgical ablation of dopaminergic fiber bundles. These lower levels of dopamine resulted in reduction in size of malonate-induced lesions. Surgical manipulation would not be treatment of choice so a pharmacological approach is desired. This study was designed to see if a compound TBZ, which is used to treat Huntington's Disease, works by reducing lesion volume in rats. Lesions were induced by injecting malonate into striatum of animals treated with i.p. injection of TBZ; control animals were untreated. Sections of striatal tissue were analyzed for lesion volume using computer-based image analysis. Results revealed that there was no significant difference in lesion volume between the TBZ treated animals and controls. While lesion reduction was the same, the lesions were much smaller by ca. 50%. It was also noted that the animals did respond to the TBZ treatment due to apparent lack of movement. This indicates dopamine was depleted. This study suggests that further research is warranted and that different dosages amounts or different dosage regimens of TBZ should be employed.

Asthma and allergies. ANNE M. CHMILEWSKI, duPont Manual High School, Louisville, KY 40208.

Louisville, Kentucky, has long been considered worse than many cities for allergies and asthma as result of several factors, including its location in a valley, its long pollen season, and its diverse plant population. A survey was designed to test the validity of considering Louisville to be an unhealthy residence for allergy and asthma sufferers. The survey, which asked questions concerning severity, longevity, and occurrence of allergies and asthma, was completed and returned by 52 students, ages 15-16, at duPont Manual High School. It was found that 63% of those surveyed had some form of allergies, compared to the national rate for the entire population, which is only about 20%. Asthma sufferers accounted for 19% of the survey population, but make up only 7% of the US population under age 18. It is also important to note that 56%,

or more than half of the students, claimed to be sensitive to tobacco smoke to some degree. Finally, non-native students (students not born in Louisville) had only a 53% allergy rate, compared to a 70% allergy rate for native Louisville residents. The experiment was greatly hampered by its limited size and range of survey participants, but the results can be seen as evidence of the need for greater study of allergies and asthma in all ages of Louisville residents.

The effects of vitamin C on runners. LISA MUDD, duPont Manual High School, Louisville, KY 40208.

Many runners spend several days recovering after hard races and practices. These recovery days often infringe on vital workouts, causing the runner to lose ground. By giving runners 500 mg of vitamin C, I hoped to determine whether vitamin C has an effect on the recovery time in the muscles of runners. Runners were each given one 500 mg vitamin C capsule each day for 5 days. Each day their flexibility was measured before they participated in a track workout. They took the weekend off, and then they did the same with a placebo. Females, being more flexible than males, noticed the smallest change in flexibility (2–4 inches). Males noticed a larger change in flexibility (2–6 inches). Females also experienced a smaller decrease in flexibility (2–4 inches), while the males experienced a larger decrease (3–6 inches).

Methods to improve body composition. JEFF KARP and MATT ROBERTS, duPont Manual High School, Louisville, KY 40208.

This project examines the impact of weight training and running on subjects' body fat percentage (BFP) and maximum weight lifts. Similar tests have been conducted before, but none dealing with this combination. To qualify for the tests, participants had to be between the ages of 14 and 18 and must adhere to the schedule set out by the researchers. The schedule required the subjects to complete a running program and a weight training session 4 times a week for 4 weeks. The running program lasted ca. 1 hr/day and consisted of form running, agility drills, distance running, and traditional sprints. The weight-training program also lasted ca. 1 hour and consisted of exercises that focused on the chest and lower body. The bench press, leg press, and squat were the three lifts used for supporting evidence. Once all of the information was gathered, it was analyzed and graphed. After running t-tests on the data we could see that there was a significant difference in the body fat percentage and the maximum weight lifts. This can be attributed to one reason: more than half of the subjects were exposed to this type of rigorous conditioning for the first time, therefore extensively improving their BFP and maximum weight lifts.

The involvement of the pro-apoptotic protein in prostate apoptosis response-4 in amyotrophic lateral sclerosis. CRYSTAL PRYOR, Nicholas County High School, Carlisle, KY 40311-9124.

Amyotrophic lateral sclerosis (ALS) is caused by degeneration of motor neurons in the spinal cord and brainstem. Familial type, which represents about 20% of all ALS cases, is caused by a gene mutation in Cu/Zn superoxide dismutase. This mutation results in an increase in oxidative stress. Recent studies have noted that the oxidative stress in familial ALS cases result in higher levels of lipid peroxidation products, which is thought to cause a type of cell death called apoptosis. A new method of detecting neuronal cell death is in measuring levels of a protein called prostate apoptosis response-4 (Par-4). This research attempted to determine if an increase in Par-4 could be detected using transgenic murine spinal cord cells containing the Cu/Zn SOD mutation, and also attempted to measure Par-4 in NSC-19 cells transfected with the mutation after treatment with lipid peroxidation products. It was expected that both cell types would demonstrate an increase in Par-4. Western blot analysis was used. Results showed that cells from transgenic murine spinal cord did have an increase in Par-4, meaning there is a connection of the protein to ALS. The transfected mutant cell line did not show an increase in Par-4 since the level of the protein was high in all cells, not just those treated. This was thought to be caused by the high rate of proliferation of the cells.

HIV causes multiple skin disorders by decreasing EGF-r density. PENG LEI, duPont Manual High School, Louisville, KY 40208.

Transgenic mice infected with the human immunodeficiency virus type-1 (HIV-1) and human patients with Acquired Immunodeficiency Syndrome (AIDS) both exhibit similar skin disorders. While the mechanism by which HIV causes such disorders is yet unclear, it is known that the skin problems and growth failures observed in the HIV-1 transgenic homozygous mice are similar to those of mice with low epidermal growth factor receptor (EGF-r) expression. Moreover, the hyperproliferative skin disorder in HIV-1 transgenic heterozygous mice is comparable to the hyperactive growth of tumors in mice with abnormally high EGF-r expression. Thus, it is hypothesized that HIV affects the expression of EGF-r in mice skin. The EGF-r densities in skin of nontransgenic and HIV-1 transgenic mice, both homozygous and heterozygous, are determined using an avidin-biotin immunoperoxidase staining technique. The data show that HIV-1 transgenic homozygous mice have EGF-r densities significantly less than those of non-transgenic mice.

Resveratrol as an estrogen agonist or antagonist in Era and Er $\beta$ . ROSEMARY SIMS, duPont Manual High School, Louisville, KY 40208.

Resveratrol is a phytoestrogen found in the skins of red grapes. It is believed by many scientists to be an explanation for the French-Paradox, the low occurrence of heart disease despite a high fat diet. This experiment was conducted to see if resveratrol would act as an estrogen agonist or antagonist in the presence of estrogen receptor

alpha and estrogen receptor beta. Prior to experimenting, the belief was held that estrogenic activity would be blocked by resveratrol in both receptors, while showing greater reactivity with ER $\beta$ . CHO-K1 cells (containing tandem copies of EREc38 DNA) were transiently transfected with the estrogen receptors, ethanol, 4-hydroxytamoxifen, estradiol, and resveratrol. After treatment, the cells were harvested and their estrogenic activity was counted in a luminometer, which expressed the values in light units of luciferase. These values were divided by the beta-galactosidase activities, leaving the fold induction of luciferase. The results showed that ER $\alpha$  was more active than ER $\beta$  in the presence of estradiol. Synergistic activity was noticed in three and four EREc38, a trend not seen in one and two EREc38. Surprisingly, resveratrol has similar activity in both ER $\alpha$  and ER $\beta$ . It is also apparent that resveratrol has stronger agonist activity with ER $\beta$  than estradiol. Due to the ability of 4-OHT to inhibit its estrogenic activity in all cases, resveratrol is proved to induce estrogen activities, therefore making it an estrogen agonist in the presence of both estrogen receptors.

Optimization of sickle cell anemia detection using PCR amplification and DdeI digestion. EFFIE WANG, duPont Manual High School, Louisville, KY 40208.

Sickle cell anemia (SCA) is caused by a point mutation in the gene of the beta chain of hemoglobin. This mutation happens to occur at a DdeI endonuclease digestion site. Therefore, while DdeI will digest the DNA from normal individuals at this site, it will not digest the DNA from SCA patients due to the mutation. Based upon this information, a scheme can be developed for a quick clinical diagnosis by amplifying the DNA harboring the SCA mutation, followed by DdeI digestion. The current study was aimed at identifying the best primer pair for PCR amplification. Human genomic DNA (normal, heterozygous, homozygous) and oligonucleotide primers were obtained from UCSEF. DNA concentrations were determined by spectrophotometry. The DNA fragment harboring the mutation was amplified using Taq DNA polymerase on a PCR cycler with the following program: denaturing at 94°C for 45 seconds, annealing at 55°C for 30 seconds, and extension at 72°C for 30 seconds. Thirty-five cycles of amplification were performed. Twelve different primer pairs were tested in the experiment. The quality of the PCR amplification were assessed using agarose gel electrophoresis. The PCR amplified DNA fragments from normal individuals. Patients were then divided into two aliquots, one of which was used for digestion by 10 U of DdeI and the other was undigested and used as a control. Both samples were then separated on a 2.5% agarose gel. Based on the results, primer pair 87N/43M<sub>2</sub> gave the best result in terms of clarity and accuracy.

The effects of tumor necrosis factor- $\alpha$  and prostaglandin E<sub>2</sub> on the expression of intercellular adhesion molecule-1 in eye microvascular endothelial cells. SUDIP K.

SAHA, duPont Manual High School, Louisville, KY 40242.

It has been established that during inflammation, Tumor Necrosis Factor- $\alpha$  (TNF- $\alpha$ ), a proinflammatory cytokine, increases the expression of Intercellular Adhesion Molecule-1 (ICAM-1) in the eye's Microvascular Endothelial Cells (MVEC), but the effects of Prostaglandin E<sub>2</sub> (PGE<sub>2</sub>) on this process are unknown. ICAM-1 is a member of the immunoglobulin gene superfamily with five extracellular Ig-like domains. The purpose of this study was to examine the effects of TNF- $\alpha$  and PGE<sub>2</sub> on the expression of ICAM-1. Two six-well chambers were cultured with MVEC. The groups that were being tested were as follows: (a) Tumor Necrosis Factor (TNF- $\alpha$ ); (b) TNF- $\alpha$  and PGE<sub>2</sub>; (c) TNF- $\alpha$ , AH23848 (PGE<sub>2</sub> antagonist), and PGE<sub>2</sub>; and (d) Mouse Serum, which contains various Immunoglobulin G (IgG). After being stained with the primary antibody (ICAM-1), the cells were examined and photographed by a Nikon high-speed fluorescence photomicroscope. The groups treated with (a) TNF- $\alpha$ , (b) PGE<sub>2</sub> and TNF- $\alpha$ , and (c) TNF- $\alpha$ , AH23848, and PGE<sub>2</sub> showed fluorescence while the group stimulated with (d) mouse serum showed none. There was no visible difference in the expression of ~ICAM-1 in the groups stimulated with (a) TNF- $\alpha$  and the group treated with (c) TNF- $\alpha$ , AH23848, and PGE<sub>2</sub>. However, the group stimulated with TNF- $\alpha$  and PGE<sub>2</sub> expressed more fluorescence than both of those groups did. These findings suggest that the expression of ICAM-1 by TNF- $\alpha$  is mediated predominantly by PGE<sub>2</sub> and that the EP<sub>4</sub> receptor (a PGE<sub>2</sub> receptor subtype) exists in eye MVEC.

Synaptology of GABA<sub>B</sub> receptors in the developing gustatory system. TONY STEFATER, duPont Manual High School, Louisville, KY 40208.

There is substantial evidence that GABAergic inhibition plays an important role in the processing of gustatory information in the rostral nucleus of the solitary tract (rNST). Previous light microscopic studies have demonstrated that GABA<sub>B</sub> receptor labeling in the rostral nucleus of the solitary tract (rNST) is present in neurons and processes. This study used electron microscopy and immunohistochemistry to determine the synaptic location of GABA<sub>B</sub> receptors in the adult and postnatal-day 10 (PND 10) rNST. Then, the sizes of the post-synaptic profiles were measured. GABA<sub>B</sub> receptors are located at pre-synaptic terminals and/or post-synaptic profiles. Labeled terminals are associated with dendrites, dendritic spines, and cell somata. Two types of GABAergic terminals in the rNST have been defined by their vesicular density: GABA-LD (low density) and GABA-HD (high density). They have a differential distribution such that GABA-LD terminals contact larger, more proximal dendrites and cell somata, while GABA-HD terminals contact smaller, more distal dendrites. I determined that when GABA<sub>B</sub> receptors are both pre- and post-synaptic label, the typical apposing dendrite has the cross-sectional dimensions of proximal dendrites and thus is likely GABA-LD. This is not true



when GABA<sub>B</sub> receptor labeling is only pre- or post-synaptic. These data suggest that the pattern of labeling for GABA<sub>B</sub> receptors may be different for the two previously defined GABAergic terminal types. They lend support to the hypothesis that GABAergic inputs to the proximal and distal dendrites may be different and reflect two GABAergic systems within the rNST capable of influencing individual neurons.

Fine motor control deficit in patients with Parkinson's Disease. JARRETT S. STULL, Nicholas County High School, Carlisle, KY 40311-9124.

More than half a million Americans suffer from the neurodegenerative disease known as Parkinson's disease (PD). The cause of PD is related to the loss of cells in the substantia nigra area of the brain, which results in a decrease in dopamine production. Dopamine, a neurotransmitter, is important for normal movement. This study was designed to determine the fine motor capabilities of PD subjects compared with normal aged subjects. As PD is associated with people usually past the age of 55, these older subjects were compared to normal aged subjects not having PD. PD subjects were tested on a fine motor control testing apparatus called the Movement Assessment Panel (MAP). It was hypothesized that the PD subjects would have a diminished response in the fine motor functions as compared to normal aged subjects. The MAP tested four tasks: removal of a nut from four shaped platforms in increasing complexity from a Platform Task to Corkscrew Task. Times for completion of the Platform Task to Corkscrew Task were measured by computer sensors. Dominant hand was measured as well as non-dominant hand. Findings revealed that the PD subjects did demonstrate the need for more time in removal of the nut in the more complex tasks, but not in the simpler tasks. These findings may hopefully lead to testing measures that will result in early detection of patients with PD.

What ocular problems does diabetes present? SOWMYA SRINIVASAN, duPont Manual High School, Louisville, KY 40208.

Twenty records of diabetic patients were analyzed to obtain relationship between diabetics vs. normal people afflicted with each of the following diseases: background diabetic retinopathy, proliferative diabetic retinopathy, macular edema, cataracts, and glaucoma. The impact of age was also studied. Data were separated by gender and then by age groups. Statistics show that after having diabetes for 10 years, 50% of diabetics will get some degree of retinopathy. After 15 to 20 years, 90% of diabetics will have retinopathy. Among this group 25% will even have proliferative diabetic retinopathy. Also, in people 40 or younger, diabetics are 25 times more likely to develop cataracts than non-diabetics. In people 60 or older, diabetics are two to four times as likely to develop cataracts; at this age people with healthy eyes already have a 50% chance of developing a cataract. Data obtained supported findings in most cases but there were exceptions due to inadequate

data for each age group. Findings show that it is inevitable for diabetics to develop some degree of ocular problems in their lifetime.

Effects of penny composition on the pH of stomach acid. MARGARET FENTON, duPont Manual High School, Louisville, KY 40208.

This study explored the relationship between the composition of pennies and the pH levels generated when they were bathed in a hydrochloric acid solution for various periods of time (1–96 hours). Pennies minted between 1962 and 1982 are a copper-zinc alloy; pennies minted after 1982 are a copper-coated zinc core. Physicians saw an increase in stomach ulcers caused by ingested pennies following the reformulation of these coins. The cause of this increased occurrence may be linked to a failure to maintain a pH balance between the gastric acids and the surface of the epithelium of the stomach. The null hypothesis states that the pH level of the simulated gastric acid will not be differently affected by the reaction rate of an "old" penny vs. a "new" penny when bathed in a hydrochloric acid solution. Based on the data, the null hypothesis was not rejected. Thus, a general change in pH is not suspected as a cause for the formation of these gastric ulcers. However, observations of rapid reactions of acid with the zinc core of new pennies, which produced hydrogen gas, suggests that local formation of hydrogen gas bubbles may be connected to the development of ulcers around pennies.

Which oils are the most healthful? SUPRAJA PARTHASARATHY, duPont Manual High School, Louisville, KY 40208.

Four different oils—olive oil, peanut oil, soybean oil, and coconut oil—were analyzed using the Thin Layer Chromatography method. Four different standards—triglycerides (bad/good, depending on the fatty acids), phospholipids (good), cholesterol (bad), cholesteryl ester (good)—were expected in the oils. These contents of the oils indicated whether the oil was healthful or not. It was concluded that peanut oil and coconut oil were unhealthy because they contained large amounts of cholesteryl ester and cholesterol. Soybean oil was healthful because it contained less amounts of cholesterol and triglycerides. The healthfulness of the olive oil could not be determined because there was some cholesterol and cholesteryl ester and lots of triglycerides. There are two different types of fatty acids in triglycerides, one good and one bad and it was not possible to determine which fatty acids were in them.

#### BOTANY

The gravitropic response of inflorescence stems of *Arabidopsis thaliana* starchless mutants. SARA R. WETZEL, Notre Dame Academy, Covington, KY 41011-2796.

The observation that a starchless mutant of *Arabidopsis thaliana* exhibits gravitropism raises questions about the hypothesis that starch and amyloplasts play a determining

role in gravity perception. I compared gravitropic response in the inflorescence stems of this starchless mutant with its wild-type counterpart. After overnight gravistimulation, I rotated the plants 180 degrees and monitored their changing curvature in response to gravity. Plants were imaged at 1-hour intervals; angles were measured by use of NIH Image software. Starchless mutant stems showed definite gravitropic responses. However, curvature of starchless inflorescence stems was markedly less than that of wild-type stems. There was a 67% difference after 6 hours in Run One and a 55% difference in Run Two. Differences in curvature were statistically significant throughout the runs. Starch is not required for gravity perception in *A. thaliana* inflorescence stems but is necessary for full sensitivity.

Which foods like yeast best? STEVEN C. EDWARDS, duPont Manual High School, Louisville, KY 40208.

This experiment was done in order to determine which type of food contained the most yeast, whether it be the dairy, bread, vegetable, fruit, or fats-and-oils food group. The procedure followed to find the yeast concentration involved forming a yeast culture and adding a measured sample of each group of food to that culture, then taking a qualitative measurement of the carbon dioxide formed. From this test it was discovered that the vegetable-and-fruit group contained the most yeast, based on the amount of carbon dioxide formed.

The effect of electrical energy on the growth of *Tagetes patula*. LAFARIN MERIWETHER, duPont Manual High School, Louisville, KY 40208.

This was an experiment to test the effect of electrical energy on the growth of *Tagetes patula*. It was assumed that the plants that received the energy treatment would grow faster than that of the untreated plants. The hypothesis was not supported by the data collected. Twelve seeds were planted. For 4 weeks all 12 plants grew on their own without any treatment. At the end of each experiment the length of each plant in centimeters was recorded. Three of the pots for the next 4 weeks were treated for 2 hours each day with electrical energy from a 6-volt battery. The electrical treatment did have an effect on the growth of *T. patula*, causing the growth process to slow down.

The effect of an electromagnetic field on the growth of *Brassica rapa*. THOMAS A. JOHNSON, duPont Manual High School, Louisville, KY 40208.

To test the effect of daily exposures to an electromagnetic field on *Brassica rapa*, a control group of 15 plants was planted in vermiculite soil and set under constant fluorescent lighting in open air. The plants were given 2 ml of water each day, and the height was recorded afterward. After the data were collected, the rates of growth were calculated. For the experimental group, the same process was done, except that, after watering, the plants were exposed to an electromagnetic field for 5 minutes set inside of a helix structure of copper wiring connected

to 6-volt battery). As a result, the rate of the control group was higher, but after a t-test was performed, the data were shown not to be significant. The rate could have been higher due to the possibility of the proteins inside the plant having a change in polarity, but, most likely, the greater rate was due to experimental error.

Plant cell mutation. JOHN MEIGOONI, Leestown Math, Science, and Technology Magnet School, Lexington, KY 40511.

Many investigators have reported human and animal cell mutations. It is interesting to know that for both humans and animals, newborn offspring are more sensitive to radiation than are adults. This is because of the fact that in children the cells divide more rapidly than in adults and hence they transfer the damages they receive to a larger number of cells. This effect appears as cell mutations. Radiation is one of the sources of cell damage, particularly in humans and animals. The goal of this project was to measure the mutation of germinated ("newborn") plant cells by radiation. This effect was measured by computing the growth of several germinated beans, which were irradiated to various doses to the growth of unirradiated beans (control). Two types of beans—kidney beans and chick beans—were used. Five beans were selected for each sample case to get a good statistical result. The beans were germinated in a wet towel. The control beans were not irradiated. Others were irradiated to doses of 1 Gy, 5 Gy and 10 Gy using a linear accelerator in the Department of Radiation Therapy, University of Kentucky. All of the beans were planted, and their growth was measured periodically. The growths of the irradiated beans were compared to the growth of the control group. The results indicated that the plant cells do not behave like human cells and that they do not show any mutation due to the radiation.

## CHEMISTRY

Determination of reactivity ratios for polyacrylate elastomers. WEI DENG and CLAYTON SMITH, duPont Manual High School, Louisville, KY 40208.

The automobile industry has recently shown an extreme favoritism towards the acrylate elastomers, namely ethyl acrylate (EA), methoxyethyl acrylate (MEA), and n-butyl acrylate (BA). Because EA has a high transition state and BA has poor oil resistance, the most auspicious combination of acrylates is that of EA and MEA. However, even with the advances in production, there has not been a record of their reactivity ratios, an essential tool used in tailor-making copolymers. Hydrogen nuclear magnetic resonance spectroscopy ( $^1\text{H}$  NMR) was used to analyze polymers synthesized in a temperature-controlled environment with bubbling argon gas. The data from  $^1\text{H}$  NMR were substituted into equations to find the reactivity ratios. The calculated reactivity ratios of EA and MEA in poly(EA co-MEA) were 0.6580 and 2.0074, respectively. In poly(EA co-VBC), the ratios were 0.0080 (EA) and 6.7180 (VBC). These numbers indicate that EA polymer-

izes three times as fast as MEA, and VBC 840 times as fast as EA. An extension of the concept applied in this project would be to investigate other reactivity ratios and develop polymerization models to aid chemical companies in creating efficient production algorithms.

#### COMPUTER SCIENCE & MATHEMATICS

A rat digital electroencephalograph: a tool for studying epilepsy. MARK J. GRUENTHAL, duPont Manual High School, Louisville, KY 40202.

Brains consist of cells called neurons. Neurons communicate by transmitting electrical currents. A device called an EEG is used to record and analyze these currents. EEGs are used mainly to detect epileptic seizures. Much testing in the study of epilepsy takes place on rats. Traditional EEGs use analog data that are displayed on scrolling paper. The problems with this are that so much paper is used and that manual analysis of the traces must be done. Through a process called analog to digital conversion, analog data can be transformed into digital data that can be stored by computer. However, these technologies have been implemented only for human EEGs and at great cost. The purpose of this project was to create a cost-efficient method for collecting digital EEG data on rats. In order to do this, software was developed to input, save, and replay EEG data with a computer. Testing was then done on rats to determine the effectiveness of the procedure.

The efficiency level of automated attendance compared to manual attendance. TRAVIS J. ACKERT, duPont Manual High School, Louisville, KY 40208.

This research has to do with the efficiency of manual attendance and automatic school attendance systems. There are many steps that go into the attendance process. Some of the steps are gathering attendance, recording those who are or tardy, distributing attendance back to teachers, and reporting absences to parents. Much wasted money is spent every year on employees and on loss of teaching time. For the experiment an automatic attendance program was created, and its efficiency was compared to that of the current automatic system. The cost, speed, and ease of use were all measured in the research. It was found that the hypothesis was supported by the research. Schools using the automated system would save more money and time than those with manual systems.

Error free web page design. MICHAEL L. BLACK, duPont Manual High School, Louisville, KY 40208.

A program was written that was intended to eliminate common errors created by popular HTML design tools. These errors include incorrect hyperlink addresses and incorrect directory structure. The program treats each item in an HTML document as a object with properties. A user would create these items, edit their properties, and adjust the order they would appear in an html document. After this the program would read through the data for each object (stored in a two-dimensional array) and use it to

fill in blanks in pre-generated HTML code. The program would also move all files to a single folder to prevent the document from having incorrect link addresses. The program also does similar tasks with style sheet documents and framesets. This software does eliminate errors caused by software; however it cannot eliminate those caused by the user.

Chaotic encryption. RYAN P. HATCH, duPont Manual High School, Louisville, KY 40208.

To encrypt messages with traditional methods, a stream of pseudo-random numbers must be generated to interact with the message according to the encryption algorithm. This project looks into the possibility of using chaotic functions to provide such streams of random numbers, thereby providing integrity to the encryption key since the chaotic functions involved are very sensitive to changes in their initial conditions. It is also very difficult to find the starting conditions of the chaotic function or to predict the next series of data from a previous series. Special emphasis has been put on the practical aspect of building a working prototype encryption system on UNIX workstations. The traditional secretly-distributed-key (SDK) approach incorporates a secret key that has to be transmitted via a secure channel to the destination. A number of keys are used in a SDK system to mix with the message in various ways to form a product cipher with the desired properties. This encryption system uses a new way to drive the key generation and coding/decoding schemes: chaotic functions. The sensitivity to initial conditions is a useful property of such functions, in providing integrity to a single key. One key will generate a properly decoded message; otherwise, the key will render the message into "garbage" only. The computer programs written include their own software floating-point library, which extends the numeric precision of the system for reasons of safety and compatibility.

#### EARTH & SPACE SCIENCE

Northern sky survey at 1420 MHz. ROBERT D. KELLY, Louisville Male Traditional High School, Louisville, KY 40228.

The goal of the research was to observe the northern sky as seen from Earth in October at 1420 MHz (atomic hydrogen band) at the Morehead Radio Telescope. After observations are taken, the data will be reduced using a reduction algorithm designed for this project. A three-dimensional map, much like a topographical map, will be compiled from this data. Much like a television builds an image on the screen through row after row of pixels, this map will be compiled by sweeping the telescope  $360^\circ$  in azimuth for every  $2^\circ$  in elevation between  $0^\circ$  and  $90^\circ$ . This will give the rows of data points, one for each sweep in azimuth. Now the data are reduced using the algorithm. This process separates the data into their respective rows by elevation, then finds the median deflection value for each tenth of a degree in azimuth. With this done, the rows are lined up in such a fashion to create a matrix,



which can be imported into a graphing utility to create the three dimensional map of the northern sky. The results of this research include a finished hydrogen distribution map of the northern sky, as well as a reusable algorithm for data reduction in future mapping observations. The map, though not as detailed as others, is a success as the first large-scale mapping observation of the Morehead Radio Telescope; it has inspired such research as mapping the entire northern sky, not just a portion of it.

Comparison of seismic activity in the areas surrounding Jefferson County and Louisville, KY. JOEL LANCETA, duPont Manual High School, Louisville, KY 40208.

A seismograph was built to test the seismic activity of the area of Jefferson County, KY. The seismograph was made from common items, such as a steel pipe, a wooden board to serve as a base, and an aluminum rod to serve as the pendulum, which vibrates at any motion. Then the seismograph was tested during December 1999 to January 2000 in an uninhabited wooded area, at a commons ground in the University of Louisville Campus ground, and at Six Mile Lane in Jeffersontown, Kentucky. The vibrations underneath were tested. A recording device was connected to the rod (pendulum) to move at any vibration. A person held one roll of paper while another pulled the paper across the seismograph touching the end of the recording device. If no movement was recorded, then the seismograph would draw only a straight line. If there were some movement, then the seismograph would draw a curved line across the paper. In two areas, the seismograph came up with a straight line. In the third area, the seismograph produced a squiggly line, followed by a straight line that remained intact. There were no reports of major seismic activity reported. Whether or not the line recorded a small amount of activity or was affected by any other variables is subject to speculation.

Erosion control: natural vs. artificial. CHRISTOPHER REZVANIAN, duPont Manual High School, Louisville, KY 40241.

Soil erosion is accelerating on a world-wide scale. What can be done about this growing catastrophe? Using a natural ditch, I tested two different methods of protecting property from erosion. Grass was grown in one section and used as a natural erosion protector. In another section of the ditch, I embedded stakes into the ground as an artificial erosion protector. These protection tests ran for 24 weeks. Results were determined by the condition of the ditch and its usefulness to control runoff. After 24 weeks of natural weathering, there was significant evidence of erosion on the majority of the ditch. At the end of experimentation, Section One, the control variable, exhibited signs of extreme erosion. In 24 weeks ca. 7.425 ft<sup>3</sup> of soil eroded. Section Three (artificial erosion control) showed very minor signs to erosion with only 1.237 ft<sup>3</sup> of displaced soil. Section Two (natural erosion control) showed signs of the least erosion, with no change in slope

and only 0.412 ft<sup>3</sup> of displaced soil. There was a minute difference in the amount of soil displaced between Section Two and Section Three in that Section Three was eroded slightly more (0.825 ft<sup>3</sup>) than Section Two. The data do not support the hypothesis that the artificial erosion-protection would defend against erosion better than the natural erosion protector. It was found that the natural erosion-protection methods proved to be more effective than the artificial methods. With support from statistical analysis, these results are substantially significant, therefore supporting the fact that the natural weathering was the only factor contributing to displaced soil in the experiment.

## ENGINEERING

Investigations of nanocrystalline solar cells. NICOLE R. CRNKOVICH, Notre Dame Academy, Covington, KY 41011-2796.

The purposes of this research were to determine whether pokeberries could be used as the photosensitive dye in a nanocrystalline solar cell, to compare its performance with raspberry-stained cells, and to study the influence of the type of conductive glass used in the electrodes. The effects of these on efficiency, emf, internal resistance, and the lifetime of the cell under open-circuit conditions were studied. This was done by constructing several nanocrystalline solar cells using raspberry and pokeberry (*Phytolacca americana*)-stained titanium dioxide on Tec 8 and Tec 15 conductive glass. Data were taken for external resistances ranging from 100 to 545  $\Omega$  and were used to determine emfs and internal resistances. Time-lapse data were also taken in open-circuit conditions, as the cells were drained. Efficiencies of each cell in converting light energy into electrical energy were calculated. Pokeberries, which are high in anthocyanin content, were successfully used, resulting in solar cells with 160 mV open-circuit voltages, compared with voltages ranging from 4 to 203 mV in raspberry-stained cells. Emfs for the pokeberry cells were in the tens of millivolts, whereas those in raspberry cells were as high as 270 mV. Internal resistances for pokeberry-stained cells were generally higher than raspberry-stained cells (100–900  $\Omega$ ). Differences between Tec 8 and Tec 15 glass were seen in internal resistances and time lapse data, with Tec 15 glass leading to higher internal resistances and cells whose performance deteriorated more rapidly. The most efficient cell achieved was a raspberry-stained cell with Tec 15 glass. This cell had an efficiency of 1.1 %.

Solar distillation of ethanol. KIMCHAU NGUYEN, duPont Manual High School, Louisville, KY 40208.

Due to the large amounts of energy utilized for the distillation of ethanol using conventional distillation methods, a more energy-and-cost efficient method is needed using solar energy. A solar still was created using a wooden case, insulation, black waterproof silicone layers, fiberglass screens, drywall siding (as the collection trough), an inlet and an outlet, and a plexiglass top, which slanted towards

the collection trough at a 20° angle. The apparatus was tested, filled with 2 liters of a 105 by volume solution of ethanol, with a lamp for 72 hours, providing 139.5 ml of distillate. The content of ethanol in the distillate could not be measured because of its small quantity, but emitted a strong ethanol odor. The ethanol distillation unit did not produce enough distillate at a high enough concentration to be considered energy efficient.

Plausibility of a hydrogen ion drive. ANDREW BUFF, duPont Manual High School, Louisville, KY 40208.

The hypothesis for this project is that the hydrogen ion engine will be fully functional and able to operate at least at 80% capacity. Last year the focus of this project was the concept and not the technology; this year the experimenter will devote all of his resources into enabling the hydrogen ion engine to work. Some of the additions made this year to improve the chances of its success are more power, a cathode/anode ionizer, and actual grids. These changes were made because it was discovered that when hydrogen is electrolyzed, it almost instantaneously combines with another hydrogen and becomes diatomic. This made the former year's method of electrostatic propulsion using hydrogen impossible; this was why a cathode and an anode were added. With the hydrogen being bombarded by electrons, it will become negatively charged, and thus the electrostatic process will be able to occur. The power output was increased in order to provide more thrust, to expedite the electrolysis process, and to strengthen the electromagnetic fields. Based on the pure mathematics and physics aspects of the project it is a complete success. While it appears to work very well in the theoretical world, the physical world is another matter. Currently, there is no accurate way of testing this type of propulsion that is not considerably expensive. However, the author of this paper will submit this theory and its schematics to NASA for review, because NASA will be able to test the idea thoroughly.

#### ENVIRONMENTAL SCIENCE

Potential use of hair to clean up oil spills. MARGUERITE BLIGNAUT, Notre Dame Academy, Covington, KY 41011-2796.

Recent NASA studies have indicated that hair may be a very useful tool in cleaning up oil spills. Hair adsorbs the oil which means the oil collects under the surfaces of the hair fibers. I used human hair in mesh bags and floated them on 10W40 oil-water mixtures. After 2 days I removed the bags and let them dry. An increase of mass indicated the amount of oil adsorbed. I found that human hair does remove oil from the water surface and that straight dark brown hair seemed to be the most efficient.

Potential use of glass tubing to enhance solar pond energy collection and storage. MICHELLE KOVARIK, Notre Dame Academy, Covington, KY 41011-2796.

The purpose of this research was to determine the effect of glass tubing on solar energy collection by a salt-

gradient solar pond. The first investigation studied the effect of a glass tubing cover on a prototype pond warming from room temperature. The second investigated the effect on maintenance of an elevated initial temperature. To carry out this research, a  $28 \times 28 \times 89$  cm tank was constructed. Temperature probes were located 6.0, 22, 52 and 80 cm beneath the water's surface, and the tank was filled with salt solutions of increasing salinity. Data were analyzed by calculating rates of temperature change at significant time intervals. The first investigation indicated that glass tubing aids in the initial warming of a solar pond and possibly later heating. For the second investigation at 10 and 30 hours, the glass tubing run showed a lower rate of heat loss. At 50 and 70 hours, the control was maintaining temperature more efficiently. Possible factors contributing to this behavior include condensation, convection, and conduction within the pond. This research indicates that glass tubing may increase the efficiency of energy collection in solar ponds, but further experimentation will be necessary before results are conclusive.

Oil absorbency of natural fibers. REBECCA S. ALLEN, duPont Manual High School, Louisville, KY 40208.

The experiment was completed in three parts: to test the fibers' oil absorption in salt water, on sand, and on concrete. To test in salt water, 450 ml of salt water and 50 g of 10W30 motor oil were placed into a container. The oil was allowed to settle for 15 minutes. Then 25 g of fiber were added and allowed to sit for another 15 minutes to absorb. The water was then separated from the oil using an oil-and-vinegar separator. The fiber was separated from the oil by using cheesecloth. The amount of oil left was weighed and recorded. To measure absorbency in sand, 200 g of playground sand were placed in a tray with 50 g of 10W30 motor oil. The oil was allowed to settle for 15 minutes before 25 g of one of the fibers were added. Next, cheesecloth was laid over the oil-covered sand. Then the fiber was added, soaking up cheesecloth oil for another 15 minutes. The cheesecloth containing the fiber and absorbed oil was then weighed, and the amount of oil absorbed was calculated. To test on concrete, 50 g of 10W30 motor oil were added to the surface and left to settle for 15 minutes. Then 25 g of one of the fibers were added and allowed to sit for another 30 minutes. The oil absorbed by the fiber was then scraped off the surface, weighed, and recorded. This process on each surface was again repeated three times for each of the four fibers. The results from the salt water found wheat bran to absorb more oil from the salt water on average. The results from the sand discovered oat bran to soak up the most oil. The final results from on concrete showed wheat bran to be most absorbent. All results were found to be contradictory to previous findings, but all were significant as proved by several ANOVA tests.

Interaction between Floyd's Fork and Pope Lick Creek (Stream Health Analysis). KUNAL KARLA\* and STEV-

EN EDLIN, Louisville Traditional Male High School, Louisville, KY 40213.

Water pollution is a major problem concerning the health of freshwater resources today. Water pollution has many sources, including urban runoff, animal wastes, sewage, and fertilizers. Controlling the amount of pollution in water is vital to our resources of freshwater, and, therefore, proper waste management techniques are essential. The purpose of this experiment was to investigate the health of two streams that are a part of the same watershed and to determine the adverse effects one stream may have on another if it is unhealthy. The area studied was where Pope Lick Creek channels into Floyd's Fork Stream. Floyd's Fork was studied upstream before Pope Lick Creek runs into it and downstream after the waters from Pope Lick Creek have been channeled. A section of Pope Lick Creek was studied as well to help determine the effect, if any, it would have on the health of Floyd's Fork. With the use of water testing, Pope Lick Creek was found to be very polluted with the most likely source of pollutants being from Nonpoint Source Pollution (NPS) because of the level of NPS pollutants found. Thus, the poor quality of Pope Lick Creek had detrimental effects on Floyd's Fork, including a poor Index of Biotic Integrity (IBI) and a poor quality of water downstream.

Soil erosion: stop the slop. CAROL ANN HAMILTON and JESSICA SCHUETTER, duPont Manual High School, Louisville, KY 40208.

Soil erosion is an enormous problem all around the globe. It may cause many detrimental problems for people in rural and even urban areas. Three variables were tested: hay, grass, and gravel. This experiment will explore the three previously listed ways in which to protect land against devastating soil erosion. Soil was placed inside a specially constructed box and was packed down tightly. Each of the three variables, one at a time, was placed over the soil, packed down tightly, and one quart of water was placed over the variable and soil, simulating rain. Each run-off was collected in a jar, labeled, and set aside. The variables, including the control, had three trials preformed and recorded. An ANOVA was run and the results of the experiment were significant at the 0.01 level. After further analysis, it was found that gravel was the best barrier from this disastrous soil dilemma, affirming our hypothesis.

#### MICROBIOLOGY

Effect of sugars on coaggregation of *Micrococcus luteus* and *Pseudomonas fluorescens*, JULIE WOLFE, Notre Dame Academy, Covington, KY 41011-2796.

Biofilms are an assemblages of microscopic animals, plants, and bacteria attached to a surface. They are of much concern in such diverse objects as industrial water pipes, dental tubing, and medical instruments. An essential step in the formation of biofilm is the coaggregation of bacteria. If the coaggregation of bacteria is inhibited, then biofilm is not able to form. Lectin-like proteins are

thought to be involved in the adhesion of the bacteria forming biofilm. Lectins often attach to carbohydrates when functioning as cell adhesion proteins, so it seems logical that sugars might affect their action. The purpose of my research was to see if adding sugar to the environment of two bacterial species expected to coaggregate would inhibit their adhesion. *Pseudomonas fluorescens* and *Micrococcus luteus* were exposed to 0.050 M solutions of glucose, sucrose, lactose, levulose, and mannitol. Absorbances were read in a spectrophotometer and percent of coaggregation for the mixture was then calculated. The calculations showed that glucose had a positive influence on the coaggregation of bacteria, while levulose had no effect. Lactose dramatically enhanced coaggregation. Results with sucrose and mannitol were inconclusive.

Evaluation of herbs as protection against ultraviolet light. ALEXANDRA MACPHERSON, Microbiology Department, Notre Dame Academy, Covington, KY 41011-2796.

The effects of ultraviolet rays on *Saccharomyces cerevisiae*, commonly known as baker's yeast, were observed in my experiment. Herbs—including rose extract, echinacea, thyme, and a combination of rose extract and echinacea—were mixed into the yeast-extract/dextrose medium. *Saccharomyces cerevisiae* strain was plated on the media. One plate of each kind was put under a germicidal UV light; one of each kind was not. In general, UV light was lethal to most yeast cells. Rose extract and thyme could possibly supply some protection from the UV light.

A reporter gene assay for methylpurine DNA glycosylase. SARA DOERR\* and ROBERT BAAR, Louisville Traditional Male High School, Louisville, KY 40243; THOMAS GEOGHEGAN, University of Louisville, Louisville, KY 40292.

The purpose of this experiment was to analyze the expression of methylpurine DNA glycosylase, a base excision repair gene. The regulatory elements of the gene were to be determined by inducing DNA damage through the use of dimethyl sulfate and examining promoter activity using a luciferase reporter gene assay. Transfection into two vectors, pCR 2.0 Blunt Vector and the pGL2 vector, was necessary. HepG2 cells were used. DMS (dimethyl sulfate) was used to treat the cells for 5 and 72 hours.  $\beta$ -galactosidase was used as a positive control for the experiment. The assay for these cells showed that the activity of the cells made a relatively small change when treated with DMS for 5 hours. Treatment for 72 hours, however, caused a dramatic decrease, which could be explained by unbalanced repair.

A transgenic construct for the cardiac-specific expression of MCAR: testing *in vivo* heart gene therapy. YAN XUAN, duPont Manual High School, Louisville, KY 40242.

A transgenic construct for the purpose of developing a mouse model for testing *in vivo* heart gene therapy. Cur-



rent gene therapy methods are ineffective due to the lack of expression of the Coxsackie Adenovirus Receptor (CAR) in non-liver organs. CAR is the receptor protein for the adenovirus, the most widely used carrier in gene therapy techniques, and is essential in the binding of the adenovirus before infection. To facilitate adenovirus transfection in cardiomyocytes, and thus make heart gene therapy more effective, a specific transgene was constructed. A murine form of the CAR gene (MCAR) was excised and cloned into a vector containing the *a-MyHC* promoter, selected because of its high transcription percentage and extreme specificity to cardiomyocytes, the target of transfection. This transgene was successfully constructed and can now be used to develop a transgenic mouse to serve as a model in testing gene therapy. This model currently does not exist, nor did a transgene utilizing MCAR for gene therapy before this study. The derived mice should have increased levels of expression of MCAR in the adult heart, thereby increasing transfection of adenovirus injected during gene therapy. Other investigators are also developing MCAR knockout mice, where MCAR is removed from all organs other than the heart, primarily the liver. The transgene developed in this study can be used independently, or in conjunction with the knockouts in development. This dual model, obviously, would be a significant improvement in gene therapy testing models.

Effect of various commonly used substances on the inhibition zones of various antibiotics on various bacteria. ANDREW B. THAI, duPont Manual High School, Louisville, KY 40208.

The purpose of this experiment was to investigate the effect of various commonly used substances on the inhibition zones of various antibiotics on various species of bacteria. It was hypothesized that, with various substances combined with various antibiotics, at least some of the combinations would exhibit the ability to serve as mutagens, by potentially altering the functional mechanism of antibiotics or chemistry of the DNA within bacteria, causing a mutation. The antibiotics and substances were tested for possible inhibition or enhancement of effects on bacteria. *Escherichia coli* and *Staphylococcus aureus*, gram positive bacteria, were chosen for experimentation. The primary technique used to collect the data was the disk diffusion susceptibility method. This method allows combinations of the substances and antibiotics to be tested on petri dishes in an even distribution manner so zones of inhibition could be observed. The antibiotic disks and substance disks could be tested together to analyze synergism. It was discovered that commonly used substances such as vitamins, pain relievers, and herbal remedies do, in fact, serve as synergistic mutagens. Also, the majority of the combinations of antibiotics and the substances have the capability to enhance inhibitory zones, therefore producing synergistic results. This synergism also has the possibility of affecting resistant rate and probability. Therefore, these results present a fresh and dynamic approach to tackling the decline of antibiotic effectiveness. The war

against resistant bacteria may not depend directly on antibiotics or bacteria, but instead on possible mutagens previously ignored that could effect the overall outcome.

Effect of curcumin as a sensitizer of radiation on human cancer cells. DAVID MEIGOONI, Leestown Math, Science, and Technology Magnet School, Lexington, KY 40511.

Curcumin is an orange crystalline powder coming from the plant *Curcuma longa*. One form of this product is commonly known as turmeric, which you may see in some foods, for example, yellow rice. Curcumin, with many clinical applications, has anti-inflammatory, anti-bacterial, and anti-tumor properties. Several investigators have published on its anti-cancerous properties. They have shown that if curcumin serum is taken orally at 500 mg/day, it prevents DNA damage caused by cancerous cells. Also, it has been shown that curcumin is an inhibitor of cancerous cells. The goal of this project is to measure the effect of curcumin as a sensitizer of radiation on PC3 prostate cancer cells. This effect was measured by comparing the survival fraction of the PC3 prostate cancer cells as a function of radiation dose with an addition of 2  $\mu\text{M}$  and 4  $\mu\text{M}$  concentration of curcumin as a sensitizer to the survival fraction of the cells with radiation alone. Enhancement of the radiation effect by the curcumin was determined as a ratio of  $\text{SF}_2$  (survival fraction for 2 Gy radiation) with radiation alone, to the  $\text{SF}_2$  with addition of curcumin. My results indicated that for 2  $\mu\text{M}$  and 4  $\mu\text{M}$  curcumin the radiation effects were enhanced by 2.9 and 17.4, respectively. These data indicate that curcumin is acting as a cell sensitizer for radiation.

## PHYSICS

Coded aperture imaging. ROSEANNE M. CHENG, du Pont Manual High School, Louisville, KY 40208.

In the field of X-ray and gamma ray astronomy, conventional image-formation devices prove inefficient in fabrication and field of view (FOV) for quick detection. Based on a pinhole camera, the efficiency of coded aperture telescopes allow fast, position detection where optimum image resolution is not essential. A simple pinhole camera makes poor use of the detector; enlarging the aperture would increase the signal-to-noise ratio (SNR) but decrease the sharpness of the image. By increasing the number of holes the source is not readily apparent seemingly due to the problem of multiple image overlap. However, if arranged according to a certain mathematical characteristic, an approximation of the original image may be obtained with a suitable computational filtering algorithm. The mask is defined over the entrance plane by the impulse response function  $h(x,y) = 1$  over a transparent area and  $h(x,y) = 0$  elsewhere. Thus, the plate can consist of a two-dimensional mosaic of transparent and opaque squares arranged such that its cyclic auto-correlation function approximates a delta function. Such a function is the characteristic of the coded aperture mask, essential for implementation in the reconstruction stage. This project

highlights the use of Wiener filtering as the optimum method for coded-masks for the ST-6 CCD camera. Implementation primarily consisted of the C programming language with strings from C++, while MATLAB was used to simulate the degradation model. The masks were designed in AUTOCAD, sent to Hamilton Printing, then appended to the ST-6.

Factors influencing the freezing hot water vs. cold water. BETH TATMAN, Louisville Traditional Male High School, Louisville, KY 40243.

Generally, one may think that cold water would freeze faster than hot water because of the laws of thermodynamics. However, many people have been known to say that hot water freezes faster than cold water yet they don't know why. The purpose of this experiment was to determine if hot water freezes faster than cold water and, if so, what are the factors that control the process. Four different temperatures were compared throughout the experiment: 20, 30, 80, and 90°C. While the first two trials compared 90°C to 30°C, the third and fourth trials compared 20°C to 80°C, and the fifth and sixth trials compared 90°C to 20°C. For each trial, the mass of both the hot and cold water as well as their ice cubes were measured to determine the amount of evaporation. Evaporation is a cooling process; additionally, it reduces the water's mass that must be frozen. When the temperature was higher, an increase in the amount of evaporation was evident. Also, the position in the freezer did not have an effect on the amount of time it took to freeze or the amount of water lost to evaporation during the cooling process. Experimentation showed that 90°C water did indeed freeze faster before the 30°C water. However the 80°C water did not freeze faster than the 20°C. Data showed that evaporation is the main cause for hot water freezing faster than cold water.

Determining the velocity at which vibrational waves travel through different waves at different harmonics. JOHN HENSON, duPont Manual High School, Louisville, KY 40208.

Every solid matter has a number of characteristic natural patterns at which it is capable of vibrating, called normal modes. Such a system is capable of oscillating at one or more than one of those normal modes or natural frequencies. When an alternating force or vibration is applied to such a system, the system is set into motion. If the force is vibrating at a frequency equal to one of the natural frequencies of the system, then the vibrations of the system are larger than if the applied force is operating at other frequencies. This is known as resonance. The waves being generated by the applied force are being reflected from the fixed ends, producing waves that are traveling in both directions. The converging wave trains together form a standing wave. From these waves, the frequency and velocity at which they are traveling can be found by using mathematical formulas. Tension applied on the material can change the harmonic at which its waves are traveling. The purpose of this experiment is to deter-

mine the velocity at which vibrational waves travel through the six different materials: cotton, copper, brass, two grades of nylon, and steel at seven harmonics. This is accomplished by hooking one end of the material to an electrically-driven vibrator and the other to a weight stand. Each material responded to the vibrating force in a unique manner. A diverse range of velocities between the materials for each harmonic was found.

## ZOOLOGY

Elasticity of the bovine aorta. LISA SOPER, Notre Dame Academy, Covington, KY 41011-2796.

A cow heart was obtained from a local meat market. The aorta was cut from the heart and sliced into rings one cm wide. The artery segments were stretched between metal rings on each end, with the top ring attached to a metal bar, and the bottom ring to a force probe. The computer interfaced probe was used to record the force as the artery was stretched and then gradually released. NIH Image was used to capture and analyze two images for each force: a far picture, showing the entire arterial ring, and a close-up. From these data, stresses and strains were calculated. The rings demonstrated elastic behavior when stretched and released. This elasticity was greatly reduced when the rings were tested after being boiled in water.

Effects of magnetic fields on maturation of the beetle *Tenebrio molitor*. MOLLIE WOLKING, Notre Dame Academy, Covington, KY 41011-2796.

The effect of magnetic fields on living organisms is still a widely studied topic. On the day of their emergence as pupae, *Tenebrio molitor* were placed in magnetic fields with strengths of 0.33 mT or 290 mT. The pupae that were placed in the 0.33 mT field were also placed in an air-conditioned room at 19°C, and the pupae in the 290 mT field were at room temperature or in a 3°C refrigerator. Pigmentation development during the first 7 days of the adult stage was studied by capturing images daily and using NIH Image software. There was no statistically significant difference in the beetles' pigmentation formation in the magnetic fields and in the control, whether at room temperature or at 19°C. The time for the pupae to emerge into adults was also studied. The average time for the pupae in the control to emerge was 16.5 days, and for the pupae in the 0.33 mT magnetic field the average was 17.3 days. There was a statistically significant difference in these values. Metamorphosis was so slowed at 3°C that adults have not emerged from the pupal stage after 40 days. Studies of respiration rates of the pupae were attempted but equipment sensitive enough to record such data was not available.

*Bacillus thuringiensis* and *Bacillus sphaericus* as insecticides against *Culex pipiens*. GREGORY WETZEL, duPont Manual High School, Louisville, KY 40208.

The spread of diseases through the mosquito vector is a problem in third-in world countries and persons who travel to these places. The fact is that most of the insect-

ticides or repellants are either harmful to the environment or not very effective. That is why there is a growing interest in formulating natural ways to repel or curb the mosquito population. This project tests the effects of two bacteria, *Bacillus thuringiensis var. israelensis* and *B. sphaericus*, as natural insecticides against mosquito larvae of the species *Culex pipiens*. There were two control groups, one containing the alga *Spirogyra*, the other void of any microbials. The first control group was utilized because if there was a difference between the mortality rate in the experimental and control groups, it would be due to the bacterium itself, not the presence of a foreign microbe. Each of the four groups consisted of 10 tubes, each with three larvae in it. One ml of broth, 1 ml of Alga-Gro, and 5 ml of distilled water were added to each tube. The groups were incubated for 120 hr at 30°C, and the number of dead larvae in each tube was recorded at 24 hr and 120 hr. The results were analyzed using an analysis of variance (ANOVA) to determine significance. The ANOVA showed the results to be significant after 24 hr, with a 0.000000165 probability that the results were random while the results after 120 hrs were shown to have a 0.57 probability that they were purely random, and thus not significant.

Effect of an antioxidant diet on the life span of *Drosophila melanogaster*. BEN WATKINS, duPont Manual High School, Louisville, KY 40208.

Antioxidants are believed to slow the aging process and extend life span by attacking free radicals, by-products of normal metabolism that cause oxidative damage. Research by Professor William Orr of Southern Methodist University showed that genetically altering fruit flies (*Drosophila melanogaster*) to increase antioxidant production almost doubled average life span. This experiment was designed to test the theory that a diet rich in antioxidants could produce similar results. The fruit flies were divided into three groups and fed three different diets throughout their different life stages. The control group was fed a basic mixture of potatoes and sugar with yeast sprinkled on top. The orange antioxidant group was fed the same basic mixture with an orange added. Oranges contain high levels of vitamin C, a known antioxidant. The tomato antioxidant group was fed the basic mixture with a tomato added. Tomatoes contain high levels of vitamin C and lycopene, and so contain two known antioxidants. Once adult flies emerged, their life spans were measured by recording the adult deaths daily. The results partially supported the hypothesis. The mean adult life span of the tomato group was highest (9.52 days), while the orange group showed a slightly lower mean adult life span (7.92 days) than the control group (8.62 days). The increased life span of the tomato group was mostly attributed to 11 of the 50 adult flies in the group. Some of these 11 flies lived almost twice as long as any individual in the orange and control groups.

*Daphnia magna*: an alternative model for *in vivo* assessment of cardiac toxicity. STEPHANIE N. GRANT, duPont Manual Magnet High School, Louisville, KY 40208.

In humans, cardiac glycosides exhibit cardiotoxic effects; some, however, are used therapeutically. *Daphnia magna* is a small invertebrate animal living in freshwater environments. The heart of *D. magna* is a small, muscular sac located dorsally behind the large eye. The purpose of this experiment was to determine if *D. magna* exhibit the same pharmacological effects as humans; if they do, then they could possibly be used as an alternative model for *in vivo* assessment of cardiac toxicity. It is hypothesized that the *D. magna* will react to cardiac glycosides the same as humans. To determine if *D. magna* exhibit the same effects as humans when exposed to cardiac glycosides, the *D. magna* were treated with different concentrations of the cardiac glycosides digoxin, oleandrin, and uzarin. Digoxin and oleandrin decrease the heart rate of humans. However, due to structural differences, uzarin does not. Ten trials were conducted, each consisting of treating 10 *D. magna* with each of the concentrations of the cardiac glycosides. The heart rates of the *D. magna* exhibited the same pharmacological effects as humans when treated with the respected cardiac glycoside. The Wilcoxon Signed Rank Test proved that the data was significant. The research hypothesis was fully supported. It was concluded that *D. magna* exhibit the same response as humans to the cardiac glycosides digoxin, oleandrin, and uzarin.

Effect of saline on the length of nymphs of the suborder Anisoptera. ZOE ZHANG, duPont Manual High School, Louisville, KY 40208.

As the population of many insects increases, an alternative form of insect control can be found in dragonflies (insects of the suborder Anisoptera). Dragonflies can eat an insect every 3 minutes, but to reach this adult form, they must spend years in the nymph stage. If the time spent in this period can be shortened, the population of dragonflies will increase, maintaining the balance of nature. In order to molt, dragonflies need copious amounts of proteins and minerals. A common mineral is sodium chloride or table salt. The hypothesis is that up to a certain threshold, the length of molting time of dragonflies will increase as the concentration of sodium chloride increases. Beyond this threshold, either the ATP of a cell will be utilized for stabilizing the intracellular environment rather than for molting, or the cells will crenate. Thirty-six dragonfly nymphs were placed in jars containing saline concentrations of 0.05, 0.1, and 0.15%. In comparison to the control (spring water), the changes in the molting rate of the nymphs show no statistically significant difference with the increase of saline concentration. Therefore, the research hypothesis is rejected and the null hypothesis that saline concentration does not have an effect on the molting rate of dragonfly nymphs must be taken into account. Because sodium chloride failed to increase the molting rate of dragonfly nymphs, it may be hypothesized that either this particular mineral, or an abundance of minerals overall, does not trigger the release of the molt regulating hormone ecdysone.



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## NEWS

The *Morehead Electronic Journal of Applications in Mathematics* (MEJAM) is a new interdisciplinary journal sponsored by Morehead State University, Morehead, Kentucky. The goal of MEJAM is to provide a refereed outlet for undergraduate students in any discipline to publish quality papers and see the results quickly. MEJAM accepts papers that are outside the realm of the typical undergraduate curriculum and that emphasize the applications of mathematics while maintaining significant mathematical interest. Papers may be historical, expository, or completely original in nature but must adhere to strict academic standards and must emphasize some aspect of the applications of mathematics. Papers from all disciplines will be considered for publication. More information about the journal and instructions for submissions can be found on the journal's website at <http://www.morehead-st.edu/colleges/science/math/mejam/>.

The Kentucky Academy of Science is seeking to complete its set of *Transactions of the Kentucky Academy of Science*. Various issues prior to 1985 are needed. Anyone willing to donate back issues or to sell them at a reasonable price should get in touch with the editor at [thiertj@nku.edu](mailto:thiertj@nku.edu).

## PUBLICATIONS

The *Sibley Guide to Birds* from the National Audubon Society is now available. This 544-page work, written and illustrated by David Allen Sibley, covers North America north of Mexico. Families and, in some cases, genera are introduced with small figures for initial comparisons. Each group is followed by individual species accounts including figures of the different forms and phases that may be exhibited by each species and also of both perching and flying views. The book has ca. 6600 illustrations (paintings) and descriptions of 810 species and 350 regional populations; it is more a reference source than a field manual. The introductory chapter briefly discusses classification and techniques of field ornithology. A second chapter discusses and illustrates the topography of birds. *Sibley* concludes with an index to common and scientific names of species. The book is a Chanticleer Press edition published in 2000 by Alfred A. Knopf, Inc.; ISBN 0-679-45122-6; \$35.00 (soft cover).

The *National Audubon Society Field Guide to Wildflowers. Eastern Region* is a revision of the first edition of the work (1979); the revising author is John W. Thieret. This 879-page book has all new photographs of 638 species, two or three per page. The introductory pages discuss the arrangement of the color plates (mostly by color and type of flower cluster), flower parts, inflorescence type, leaves, and plant classification and names. For each included species the text gives a description, flowering times, habitat, range, and comments. Relevant families are briefly discussed. The book concludes with an index to common and scientific names of species. The book is a Chanticleer Press edition published by Alfred A. Knopf, Inc., in 2001; ISBN 0-376-40232-2; \$19.95 (soft cover).

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